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Experiments on Wool from Copper-Deficient Sheep

Part I: Some Physical Measurements on Intact Fibers and Experiments on the Dissolution and Fractionation of Oxidized Wool

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Abstract

It has been confirmed that wool from copper-deficient sheep has a faster rate of extension in water under constant load than normal; it has been found that this wool has a greater diameter swelling in formic acid and is more easily penetrated by certain large reagent molecules than normal.

The dissolution in aqueous buffer solutions of copper-deficient and normal wool after oxidation has been examined and further evidence that the copper-deficient wool is deficient in a sulfur-rich protein fraction, whose absence may be responsible for its physical abnormalities, has been obtained.

Introduction

Because of its large and unmistakable physical abnormalities, copper-deficient wool[1] is in many ways especially suitable for experiments on the interpretation of the physical properties of wool samples in terms of the structure and chemistry of the fibers. Investigations in this laboratory have therefore been aimed at a complete structural or chemical explanation for the abnormal behavior of copper-deficient wool. Some aspects of this work, including further details of tests on the role of the sulfur-containing amino-acid residues in the extensibility of the fibers, new tests on fiber diameter swelling and reagent penetrability, and more experiments on the dissolution of normal and copper-deficient wool after oxidation are given here.

From the work of Marston and others [21–25] it is now well established that copper is essential for the nutrition of sheep and that an early effect of copper deficiency is that an abnormally long time is required for keratinization in the wool follicles. Probably as a result of this, copper-deficient merino wool has abnormal crimp which may, according to the degree of deficiency, be entirely absent or consist of large irregular waves or of large waves superimposed on the normal small-amplitude crimp. Copper-deficient merino wool has a low tensile strength, different dyeing properties, and impaired elasticity when compared with normal wool. It also

[1] For convenience wool from sheep reared on a copper-deficient diet is referred to here as "copper-deficient wool." This wool actually has less copper than normal [10], although the amounts in the fibers are too small to affect their properties.

has a slightly smaller average fiber diameter, and it processes badly [22, 24, 28].

In 1946 Marston [22] concluded that the differences revealed by tests on copper-deficient and normal wool from the same sheep (up to 15% less sulfur in the copper-deficient wool but little difference in the non-sulfur-containing amino acids nor in the x-ray diffraction pattern) were not enough to explain the remarkable behavior of copper-deficient wool. It is certain that a variation of 15% in sulfur content may occur among wools that have never experienced copper deficiency without such a spectacular change in physical properties, but it has since been shown [9, 28] that copper-deficient wool contains more sulfhydryl groups than normal so that the sulfur content is not a good measure of the number of disulfide groups on which, ultimately, most of the properties of the fiber would be expected to depend. In some copper-deficient samples examined in this laboratory, sulfhydryl groups account for nearly 10% of the sulfur, and there are nearly 30% fewer disulfide groups than normal. No other comparable variations in composition between the two sorts of wool have been found. Copper-deficient wool apparently has a higher concentration of N-terminal residues than normal [10], but the significance of this is not clear, especially since Thompson's work on N-terminal residues in wool and wool fractions [33]. In spite of its abnormal crimp, copper-deficient wool has the same bilateral structure as normal according to tests in this and other laboratories. No difference between the two sorts of wool in the extent of hydrogen–deuterium exchange [7] has been found (unpublished result).

The difference in the sulfur-containing amino-acid concentrations is thus the only large chemical difference between normal and copper-deficient wool that has been found so far, and it is important to determine whether this can account directly for the physical differences. It is also of interest to determine how the structure of the fiber is altered by the absence of a large proportion of the disulfide groups. No conclusive solutions can yet be given to these problems. Experiments described here on one aspect of the extensibility of wool fibers of both sorts suggest that the extra sulfhydryl groups and deficit of disulfide groups in copper-deficient wool can largely, but probably not entirely, explain their differences in behavior. Experiments on peptides in tryptic digests of oxidized normal and copper-deficient wool, to be reported later, suggested that the copper-deficient samples contained less of a small group of highly acidic, sulfur-rich peptides, although no other differences could be found. This led to measurements [11], further details of which are given here, on the proportions of some of the protein components in each wool. It is very likely from these experiments that copper-deficient wool contains less of a sulfur-rich protein fraction than normal wool, which suggests that the missing disulfide

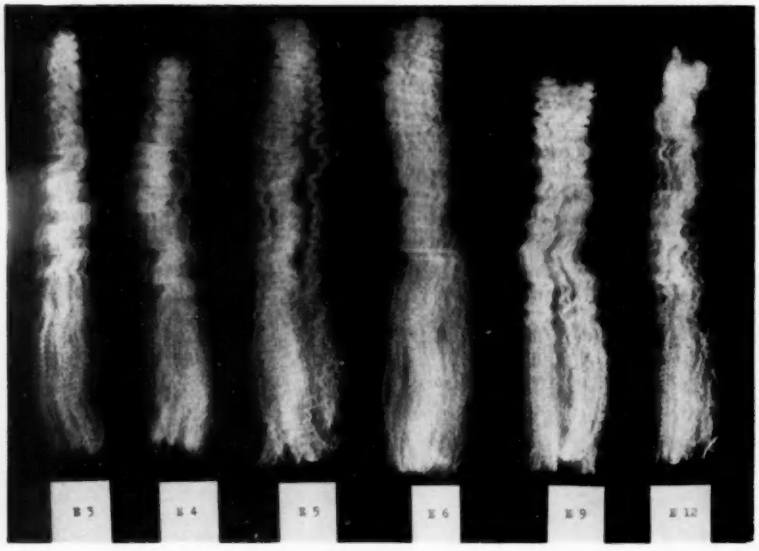

Fig. 1. Experimental wool staples; lower parts normal, upper parts copper-deficient.

groups in copper-deficient wool were attached to a particular fraction of the wool protein and were not distributed randomly.

Materials and Methods

Wool Samples

In experiments such as those reported here, it is possible to eliminate the natural variations among wool from different sheep by using fibers from one sheep that are half normal and half copper-deficient. For this purpose sheep with a long growth of wool that is known to be copper deficient are fed a supplement of cupric sulfate. The nature of the wool then changes, over a distance of a millimeter or less, and the sheep are shorn when enough "normal" wool has grown. This procedure was used in the early experiments in Australia [22] and in South Africa [30], and the merino samples used for the tests reported here were grown in the same way [10]. Figure 1 shows staples from the six experimental sheep. The demarcation between the two sorts of wool is very clear. The only differences between the growth of these wool samples and those grown in earlier experiments are that the sheep were kept partly covered with canvas patches to minimize damage to the fibers during growth and that the sheep were moved to another locality when the copper supplement was started so that the rest of the diet was not exactly the same. Other dietary changes can affect the sheep's wool, but not in such a spectacular manner as copper deficiency. Tests were usually done on samples from other merino sheep that had never experienced copper deficiency in order to be sure that the "normal" parts of the experimental staples did not deviate from the usual range of values for merino wool. Some tests were also done on staples from sheep suffering from very severe copper deficiency that were used in earlier experiments. These staples were kindly provided by Professor Swart.

For most experiments, staples from the back of each experimental sheep were divided into copper-deficient and normal segments and then purified as usual [8]. The exact division between the two sorts of wool is easier to distinguish before the fibers have been washed in water. Experiments were done on staples from within a few square inches of skin because different regions of the skin of these sheep were apt to produce fibers of slightly different appearance. An unusual feature of some of the copper-deficient fleeces, which has not been studied but is probably an example of mosaicism, was the presence of islands a few square inches in area of well-crimped fibers surrounded by the almost crimpless fibers of the rest of the fleece.

For single-fiber measurements the staples were washed in solvents as usual [8]; then fibers were selected, at random as far as possible, numbered, divided into the two parts, and washed in water. Tests were always done on both parts at the same time, although the results for several fibers were usually pooled.

Buffer Solutions

The pH 6.0 buffer used for most dissolution experiments contained 1% of pyridine and 0.1% of acetic acid by volume. Of the buffers used for Figure 6, that of pH 5.1 contained 1% of pyridine and 1% of acetic acid; those between pH 8 and 10 contained sodium bicarbonate or carbonate or mixtures of these, the strength being 0.12 *M*; those above pH 10 were ammonia solutions. A 1% pyridine solution and a 3% collidine solution were also used. They gave wool protein solutions of pH 6.7 and 8.3 respectively.

The Measurement of Fiber Diameter Swelling

The usual method was adopted; the normal and copper-deficient parts of three or four fibers were mounted on a microscope slide and the diameters measured in air at millimeter intervals using a microscope with graduated eye-piece, 30 or more measurements being taken on each fiber. The slide was then flooded with the appropriate liquid, covered with a cover glass, and the diameters remeasured at about the same places after the fibers had been allowed to equilibrate.

The Oxidation of Wool Samples

The wool samples (0.100 g. of known moisture content) were left at 5° C. with 2.5 ml. of a performic acid mixture. This was prepared by mixing one volume of 30% hydrogen peroxide with nine volumes of 96–98% formic acid and leaving the mixture to stand at room temperature for an hour [34]. After the desired period of oxidation, usually 4.5 hr., the acid mixture was sucked off under oil-pump vacuum. Most of the liquid was removed in 15 min. and the fibers were dry in 30–60 min. The

sample was left under vacuum over sodium hydroxide flakes for 24 hr. or more to remove traces of acid.

The Dissolution of Oxidized Wool Samples

The oxidized wool sample (0.100 g.) was shaken at 30° with 20.0 ml. of the appropriate buffer solution, usually for 6–7 hr. The oxidation and dissolution were done in the same flask, a 50-ml. glass-stoppered conical flask being most convenient. The undissolved material was filtered off with gentle suction through a medium-porosity sintered glass crucible and weighed after being dried for 16 hr. at 103°. The filtrate was usually clear.

The Folin-Biuret Method for Measuring Protein Concentration

The concentration of wool protein in solution was sometimes measured by the colorimetric Folin-biuret method as developed by Lowry and others [20]. To 1.0 ml. of solution, containing from 10 to 100 μg. of wool protein, 5.0 ml. of the alkali-copper reagent was added. After 5 min., 0.5 ml. of the Folin-Ciocalteu reagent [15] was added; the color intensity of the resulting blue solution was measured after 20 min. by means of a "Spekker" absorptiometer with No. 608 red filters.

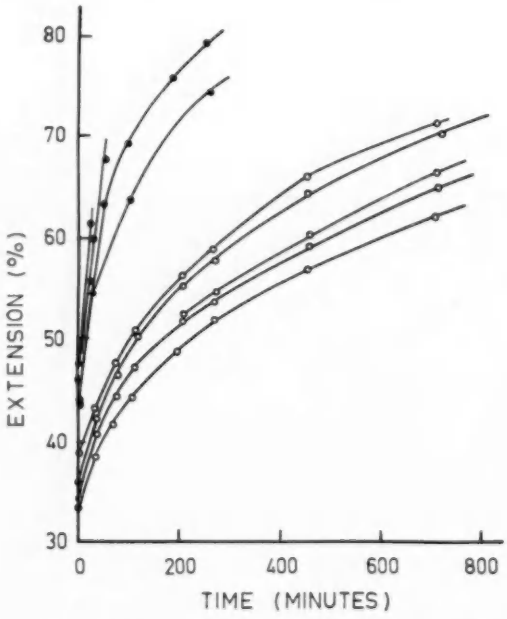

Fig. 2. Extension-time curves for copper-deficient fibers (upper curves, full circles) and normal fibers (lower curves, open circles) from sheep E3 in water under conditions described in text. Some of the normal fibers reached 80% extension after 2000 min.

Experiments and Results

The Extensibility of Copper-Deficient Wool and the Role of Sulfhydryl and Disulfide Groups

One manifestation of the abnormal physical behavior of copper-deficient wool is its faster rate of extension under constant load in water. Figure 2, for example, shows the extension of normal and

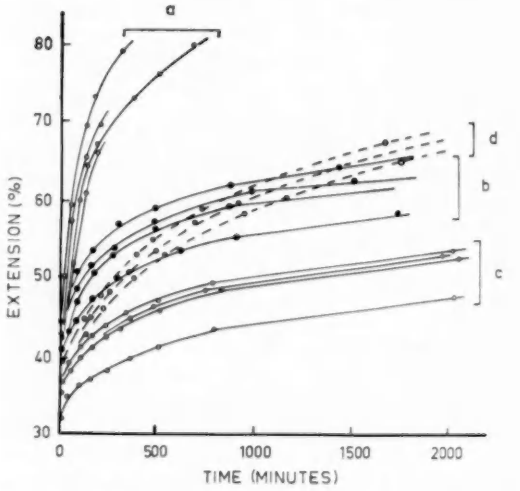

Fig. 3. Extension-time curves for fibers from sheep E9. (a) untreated copper-deficient fibers; (b) copper-deficient fibers after N-ethylmaleimide treatment; (c) normal fibers after this treatment; (d) untreated normal fibers.

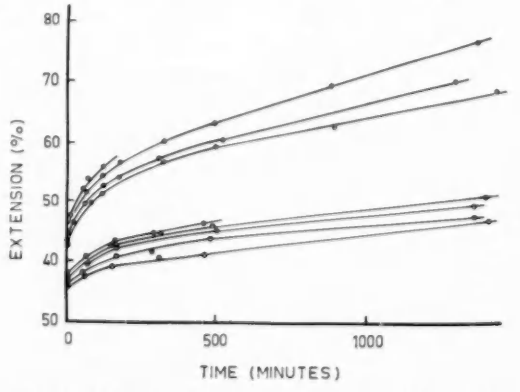

Fig. 4. Extension-time curves for copper-deficient (upper curves) and normal (lower curves) fibers from sheep E9 stretched in 0.1 *N* hydrochloric acid.

TABLE I. Values for Initial Extension (E_1) and Cystine Content of Normal and Copper-Deficient Fibers

Wool from sheep	E_1, %			Cystine		
	Normal	Cu-def.	Difference	Normal, μmoles/g.	Cu-def., μmoles/g.	Difference, %
E3	35.4	45.6	22.4	500	357	28.6
E4	34.3	37.2	7.8	533	458	14.1
E5	33.8	40.5	16.5	528	442	16.3
E6	35.8	44.5	19.6	453	390	13.9
E9	35.0	42.6	18.0	522	408	25.3
E12	35.0	39.0	10.3	459	403	12.2

copper-deficient fibers from sheep E3 measured in water at 25° under a load of 6×10^5 g./cm.^{2} by the method of Burley and Speakman [4]. Similar results were obtained with the other five pairs of experimental samples.

Extension–time curves consist of two parts: the initial instantaneous extension (E_1), measured for convenience after 1 min., and the slow extension or creep which continues until the fiber breaks [32]. The rate of the slow extension is greatly affected by the pH of the solution and by the concentration of sulfhydryl groups in the fiber, and it has been postulated that sulfhydryl–disulfide interactions occur during this stage [5, 6]. Sulfhydryl groups do not, however, greatly affect the instantaneous extension.

In order to compare the effect of sulfhydryl groups on the extension–time curves of normal and copper-deficient wool, fibers of each sort that had been treated with N-ethylmaleimide to remove sulfhydryl groups quantitatively [9] were stretched in water as usual. To examine the effect of the pH of the solution, untreated fibers of each sort were stretched in 0.1 *N* hydrochloric acid.

Figure 3 shows extension–time curves for copper-deficient fibers from sheep E9 with and without N-ethylmaleimide treatment (Groups b and a) and those for the corresponding normal fibers (Groups c and d). Curves for copper-deficient and normal fibers stretched in acid solution are shown in Figure 4. The groups of curves in Figures 3 and 4 show that, as in the case of normal fibers, N-ethylmaleimide treatment has a profound effect on the rate of extension of copper-deficient fibers, presumably owing to the removal of sulfhydryl groups, and that, as usual, this is paralleled by the effect of an acid solution on the extension of untreated fibers.

The groups of curves in Figure 3 are reminiscent of a series of curves for non-copper-deficient merino fibers [6] that had first been treated with N-ethylmaleimide to remove the naturally-occurring sulfhydryl groups (cf. Group c), then with bisulfite to produce more sulfhydryl groups and decrease the concentration of disulfide groups (cf. Group a), and finally with N-ethylmaleimide again to remove the new sulfhydryl groups without affecting the disulfide groups (cf. Group b). This suggests that the abnormally high extension–time curves for copper-deficient wool could be explained qualitatively by the increased sulfhydryl-group concentration and decreased disulfide-group concentration in this wool. It is, however, difficult to determine whether these differences are enough to explain the abnormal behavior of copper-deficient wool quantitatively. According to Groups b and c in Figure 3, N-ethylmaleimide treatment probably has a greater effect on the copper-deficient than on the normal fibers; this is consistent with the greater sulfhydryl-group concentration in the former—80 μmole/g. compared with about 20 μmole/g. The two groups are very similar if allowance is made for the higher initial extension of Group b. It is therefore possible that the difference in the second part of the extension–time curves of the untreated normal and copper-deficient fibers may be largely explained by the difference in sulfhydryl-group concentrations. Complete coincidence of Groups b and c would not be expected because the initial extension (E_1) is not so dependent on N-ethylmaleimide treatment.

In Table I the average E_1 values and the cystine contents of normal and copper-deficient fibers from each of the six experimental sheep are given. The cystine contents were obtained by subtracting the sulfhydryl-group concentration, measured by a direct method [8, 9], from the cystine plus cysteine concentration, measured by the phospho-18-tungstic acid method as used by Park and Speakman [29]. Table I shows that the copper-deficient fibers always have

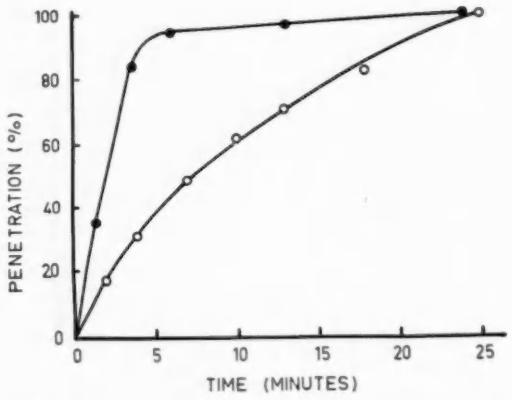

Fig. 5. Rate of penetration of 1-(4-chloromercuriphenylazo)-naphthol-2 in formamide solution into wool fibers. Full circles, copper-deficient fibers; open circles, normal fibers. Wool E6.

a higher average E_1 value than normal and that there is a tendency for large differences between the E_1 values for copper-deficient and normal fibers from the same sheep to be accompanied by a large difference in cystine content. There is not, however, a large correlation between difference in E_1 and difference in cystine content ($r = 0.68$). Thus, although there is an indication that the decrease in disulfide-group concentration is responsible for the difference in the initial part of the extension–time curves, these results suggest that other factors may be important as well.

Tests on the Accessibility of Sulfhydryl Groups and on Fiber Diameter Swelling

During studies on sulfhydryl groups in wool, reported already [8], it was found that the sulfhydryl-group reagent 1-(4-chloromercuriphenylazo)-naphthol-2 in solution in formamide penetrates copper-deficient wool more rapidly than normal wool. The rate of reaction of this reagent with sulfhydryl groups in disintegrated wool is rapid, so that the apparent rate of reaction with these groups in intact wool may be used as a measure of the rate at which the reagent penetrates the fibers. This is done in Figure 5, which shows the rate of penetration of the reagent, expressed as the percentage of the total number of sulfhydryl groups that has reacted at a given time, into samples of normal and copper-deficient wool from sheep E6. It is clear that the copper-deficient fibers are more easily penetrated by the reagent. Sulfhydryl-group reagents with small molecules do not apparently show such a large difference. It has already been shown [9] that N-ethylmaleimide in aqueous solution penetrates and reacts with the sulfhydryl groups in normal and copper-deficient wool at about the same rate.

One reason for the increased accessibility of the sulfhydryl groups in copper-deficient wool to large reagent molecules could be a greater diameter swelling of these fibers. It is difficult to measure fiber diameter swelling in anhydrous formamide because it absorbs water rapidly, with an alteration in some of its properties. Measurements can, however, easily be done in formic acid, and it was found that copper-deficient fibers swell to a significantly greater extent than normal in 97% formic acid, as shown in Table II. No difference could be found between the diameter swelling of the two sorts of wool in water or in 0.1 *N* hydrochloric acid, possibly because swelling in these media is relatively small and errors of measurement large.

Greater fiber diameter swelling and increased accessibility to reagents would perhaps be expected from the lower concentration of disulfide groups in copper-deficient wool, although, once again, it would be difficult to be certain that this alone was responsible for the difference between the copper-deficient and normal fibers.

The Oxidation, Dissolution, and Fractionation of Normal and Copper-Deficient Wool

The separation of wool keratin, after oxidation or reduction, into fractions with different properties and chemical composition has been an active subject for research since the experiments of Goddard and Michaelis in 1934 [17], although the problem of

TABLE II. Diameter Swelling of Normal and Copper-Deficient Fibers in 97% Formic Acid

Wool	Diameter swelling, average, %	No. of fibers	Coeff. of variation, %	Standard error
E5 Normal	55.9	8	3.0	0.6
Cu-def.	60.5	8	3.5	0.8
E6 Normal	54.3	6	7.8	1.9
Cu-def.	57.5	6	4.0	1.0
E9 Normal	46.8	6	6.8	1.4
Cu-def.	54.1	6	6.5	1.6
E12 Normal	52.3	6	7.1	1.9
Cu-def.	57.9	6	5.3	1.5

estimating and eliminating unwanted degradation during treatment has not yet been solved satisfactorily [19, 35, 16, 27]. After oxidation a large proportion of wool fiber protein may be readily dissolved at high pH, and, although a major component may be precipitated by acidifying the solution, a sulfur-rich minor component does not precipitate [1, 13, 14]. This seemed the most suitable method for fractionating wool proteins, so experiments have been done on normal and copper-deficient wool in an attempt to compare the proportions of these components. First the dissolution of oxidized samples of the two sorts of wool in solutions of different pH was examined.

Samples from sheep E5 that had been oxidized for 4.5 hr. in performic acid were shaken with aqueous buffer solutions of from pH 5 to 11 according to the procedure given under "Methods." After 6–7 hr. the undissolved wool was filtered off, washed with water, dried, and weighed. The amount dissolved was calculated as a percentage of the dry weight of the original wool, a small correction being applied for the increase in weight due to oxidation. Between two and six tests were done at each pH and average values were used to plot the curves in Figure 6, which shows the dissolution of oxidized normal and copper-deficient wool as a function of pH. The pH recorded is that of the equilibrium solution which with weak buffers sometimes differed slightly from that of the original buffer.

It is clear from Figure 6 that, as expected from previous work, the dissolution of each wool increases with increasing pH, but that, unexpectedly, over a wide pH range the normal wool is *more* soluble than the copper-deficient wool. The points in Figure 6 represent amounts of wool dissolved after shaking with buffer solution for 6–7 hr., during which time only the solutions of pH 6 and less had reached equilibrium. After a longer time the higher pH

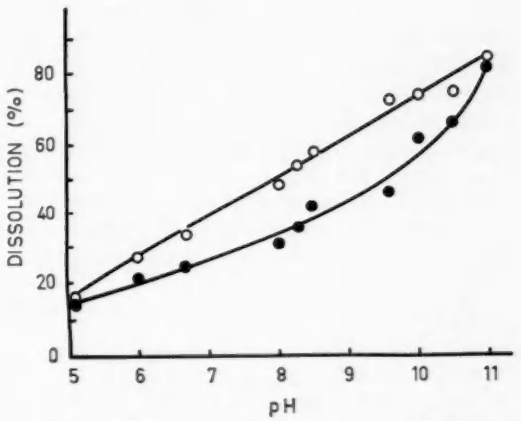

Fig. 6. The dissolution of oxidized wool E5 in buffers of different pH. Full circles, copper-deficient wool; open circles, normal wool.

buffers dissolved more protein. The pH 8.3 buffer, for example, dissolved 64.4% of the oxidized normal wool and 45.8% of the copper-deficient wool in 25 hr., and equilibrium had still not been reached.

In view of the complexity of the results in Figure 6, an attempt was next made at comparing the amounts of precipitate formed on acidifying wool protein solutions of high pH. Difficulty was, however, experienced in obtaining consistent results, and it was found that the ionic strength of the solution is important. The following procedure was used: after the oxidized wool sample had been shaken with the appropriate solution for 6 or 7 hr. and the undissolved material filtered off, a portion of the filtrate (5.0 or 10.0 ml.) was acidified to pH 3.2–4.0 with acetic acid. The mixture was allowed to stand for 16 hr. and the precipitate then centrifuged or filtered off, washed in 0.1 *N* hydrochloric acid, dried, and weighed. Table III shows some results from this method, including tests on ammonia solutions to which sodium chloride had been added to increase

TABLE III. Dissolution and Acid Precipitation of Oxidized Normal and Copper-Deficient Wool from Sheep E5

Solution	Final pH	% of original wool					
		Insoluble		Acid soluble		Acid precipitate	
		Normal	Cu-def.	Normal	Cu-def.	Normal	Cu-def.
0.5 *M* NH_4OH	11.0	15.7	18.7	35.8	31.6	48.5	49.7
0.2 *M* NH_4OH	10.8	19.6	24.9	26.9	24.5	53.5	50.6
0.2 *M* NH_4OH + 0.1 *M* NaCl	10.7	27.8	33.6	37.4	36.2	34.8	30.2
0.2 *M* NH_4OH + 0.2 *M* NaCl	10.8	25.4	34.6	41.5	34.1	33.1	31.3
0.2 *M* NH_4OH + 0.3 *M* NaCl	10.7	37.6	43.6	39.8	37.6	22.6	18.8

the ionic strength. The amounts of the dissolved material, the precipitate on acidifying, and, by difference, the acid-soluble material, are given as percentages of the original dry wool, each figure being the average of two or more measurements. The wool samples were, as before, normal and copper-deficient parts of fleece E5.

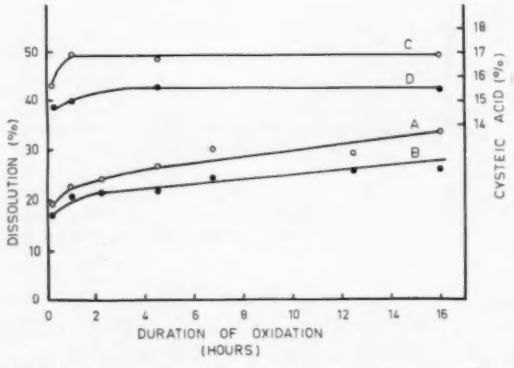

Fig. 7. Relation between duration of oxidation and (i) dissolution in 6 hr. at pH 6 (left-hand axis, lower curves, (A) normal; (B) copper-deficient wool); (ii) cysteic acid concentration (right-hand axis, upper curves, (C) normal; (D) copper-deficient wool). Wool E5.

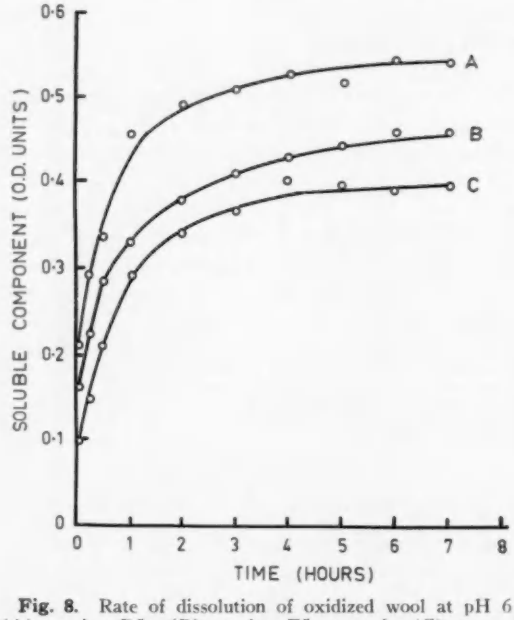

Fig. 8. Rate of dissolution of oxidized wool at pH 6: (A) merino D5; (B) merino E5, normal; (C) copper-deficient. Ordinates soluble component, i.e., dissolved protein, optical density units after Folin-biuret test.

It is apparent from Table III that the amount of material dissolved initially and the amount of precipitate are dependent on the ionic strength of the buffer, in agreement with observations of Corfield, Robson, and Skinner [12] that the dissolution of oxidized wool depends on the type of buffer. This complication does not, however, conceal the fact that the copper-deficient wool always has the *lower* solubility and *less* of the acid-soluble material, although it makes it difficult to determine the absolute amounts of the different components by this method. In order therefore to compare a large number of wool samples, a method, based on the results in Figure 6, that was empirical but simple to apply and reproducible, was adopted: the oxidized sample was shaken with the pH 6 pyridine-acetate buffer and the undissolved material (referred to as the "insoluble component") was separated and weighed as usual. The amount of the "soluble component," which went into solution, was obtained by difference. Amino-acid analyses and other tests on this soluble component, to be reported in Part II, suggested that it consists chiefly of the sulfur-rich protein fraction. Thus, by this method a rapid separation of oxidized wool into components with different amino-acid composition was achieved. It was found that at pH 6 the extent of dissolution was less influenced by the nature and ionic strength of the buffer than at pH 11.

Before comparison of other wool samples, the effect of the oxidizing process on the results was examined. In Figure 7, Curves A and B show the effect of duration of oxidation in performic acid under the usual conditions on the amounts of the soluble component obtained from normal and copper-deficient wool from sheep E5. On the same graph the cysteic acid concentrations of normal wool (Curve C) and copper-deficient wool (Curve D) are given. These analyses will be described in Part II. It is clear from Curves A and B that the difference between the normal and copper-deficient wool in the amount of the soluble component is reasonably independent of the duration of oxidation, although in each case the amount dissolved increases slightly with increasing duration of oxidation. Oxidation for 4.5 hr. was used for most experiments. The cysteic acid analyses show that oxidation of each sort of wool is complete in 4 hr. or less and that, as expected, the oxidized copper-deficient wool has less of this amino acid.

To return to the comparison of the amounts of

the soluble component in wool from sheep other than E5: Table IV gives results for (i) the normal and copper-deficient wool from the six experimental sheep; (ii) wool from five merino sheep that had never experienced deficiency of copper; (iii) wool from four merino sheep in an advanced stage of copper-deficiency—some of them with "straight steely" wool; and (iv) two non-medulated Lincoln wool samples and a non-medulated sample of Cape kid mohair. These results are all expressed, as before, as the amount of soluble component as a percentage of the original dry wool. A correction, to be discussed below, was applied for the increase in weight of the insoluble component on oxidation. Some of the figures in Table IV were used in calculating the ratios given in a preliminary communication [11].

Table IV shows that, for each of the experimental merino fleeces, the copper-deficient wool has a lower percentage of the soluble component than normal. Most of the copper-deficient samples have about 15% less of this component, although E9 is anomalous in having only 4% less in spite of a large difference in properties between the normal and copper-deficient parts of this fleece. As would probably be expected, the Lincoln wool samples, which were apparently not copper-deficient, are not comparable with the merino samples. They differed both from these and from mohair.

In making comparisons among different wool samples it is necessary to consider the correction applied for the increase in weight on oxidation. For this purpose a factor of 5% was subtracted from the weight of the insoluble component. A discussion of the derivation of this factor is given in Part II, where it is shown that a 5% correction is best for wool E5. A difference in sulfur content between the insoluble components of two wool samples will alter the weight change on oxidation and therefore the apparent proportions of the two components. It is, however, shown in Part II that, in spite of the different disulfide-group concentrations in the normal and copper-deficient fibers from sheep E5, there is no difference in the cysteic acid concentra-

TABLE IV. Protein Dissolved from Oxidized Wool Samples at pH 6

Sample from sheep	Soluble component							
	Normal wool				Copper-deficient wool			
	Av.,* %	No.*	CV,* %	SE*	Av.,* %	No.*	CV,* %	SE*
(i)								
Merino E3	28.6	4	5.0	1.2	22.2	5	6.7	0.7
E4	27.5	4	4.8	0.8	23.5	3	5.1	0.8
E5	26.7	8	7.9	0.8	21.6	8	7.0	0.6
E6	31.2	4	8.3	1.5	26.7	3	3.7	0.7
E9	25.1	8	8.1	0.8	24.1	7	10.1	1.0
E12	29.8	6	7.4	1.0	26.2	7	7.3	0.8
(ii)								
Merino D5	30.2	8	9.9	1.1				
Hr1	29.3	4	1.0	0.2				
N8	29.7	4	10.2	1.8				
N1	29.5	2						
22	29.5	9	6.2	0.6				
(iii)								
Merino S65					16.9	3	4.2	0.5
S57					19.9	4	9.7	1.1
S67					21.5	2		
S61					25.7	2		
(iv)								
Lincoln 1	21.4	2						
2	18.2	2						
Mohair	30.8	5	12.9	2.0				

* Av. = average; No. = number of estimations; CV = coefficient of variation; SE = standard error.

tion of the insoluble components. Consequently, since the weight increase is largely determined by the conversion of cystine to cysteic acid, the magnitude of the correction factor cannot greatly affect the comparison of results for the normal and copper-deficient parts of this fleece. Presumably this also applies to the other experimental fleeces.

In order to be quite certain that the correction factor necessary for the above method was not responsible in any way for the different results for normal and copper-deficient wool samples, further evidence was sought by measuring directly the amount of the soluble component in solution by means of the Folin-biuret test (see "Methods"). Figure 8 shows the rate of dissolution of equal weights of oxidized wool, normal and copper-deficient, from sheep E5 and normal wool from sheep D5, in pH 6 buffer at 30°. It is difficult to obtain the actual amounts of dissolved protein by this method, so protein concentrations are given in optical density units. Nevertheless, the equilibrium values in Figure 8 place the relative amounts of the soluble components in these samples in the same order as given in Table IV.

The effect of the action of the weather (sunlight, dust, etc.) on the amount of the soluble component that could be obtained from a merino fleece after oxidation was examined by measuring the amounts of this component in the tip, middle, and root portions of a non-copper-deficient fleece. Such measurements are important because the less compact copper-deficient fleeces are very liable to damage by weathering. The tip and middle portions were found to have a slightly lower proportion of the soluble component than the root portion, but the difference was not significant and all the values were higher than any for the copper-deficient fleeces.

Discussion

The above results show that copper-deficient wool has a faster rate of extension under constant load in water and greater fiber diameter swelling in formic acid than normal wool and that it admits certain large organic molecules more easily. These are properties that would be expected from the decreased disulfide-group concentration or from the increased sulfhydryl-group concentration in copper-deficient wool, although it is unlikely that they are solely responsible.

Whether or not the sulfur-containing amino acids are largely responsible for the abnormalities of copper-deficient wool, the problem as to where the missing disulfide groups in this wool originated and how they came to be missing is of particular interest. There are many possibilities. For example, some half-cystine residues might be missing at random along all the protein chains in the copper-deficient fibers, or only those associated with a particular amino-acid sequence might be missing, or those in a particular protein fraction might be missing. The above results for the dissolution and fractionation of oxidized fibers definitely implicate a sulfur-rich protein fraction, and it is suggested that the decrease in the concentration of disulfide groups in copper-deficient wool is really a manifestation of the fact that this wool has a lower proportion of a protein fraction that is very rich in cystine residues. If this is true, and if this fraction acts as an "intermolecular cement" between other protein chains or groups of chains [11], then some of the properties of copper-deficient wool become easier to understand, especially those that do not appear to depend solely on the concentration of disulfide groups. Thus, fibers of this wool would be weaker and more easily penetrated by reagents not only because of a decrease in the number of disulfide groups but also because of a decrease in the amount of a structurally important protein fraction. It is also possible that there may be an opportunity for sulfur-rich material to be lost to the fiber during the prolonged zone of keratinization in copper-deficient wool, especially if this material is of low molecular weight. The high concentration of sulfhydryl groups in copper-deficient wool would fit naturally into this scheme if the extra groups in this wool were potential points of attachment for the missing protein fraction. A measurement of the distribution of sulfhydryl groups in copper-deficient wool, which is now being attempted, may give evidence on this subject.

It is necessary to examine closely the assumption that copper-deficient wool is deficient in a certain sulfur-rich protein and that this is at the basis of its abnormal behavior. A number of independent techniques led to the conclusion that the protein in the cells of the more important keratinous structures, such as the wool and hair cortical cells, consists of two or more distinct proteins. In 1933 Astbury and Woods [2] from fiber extensibility experiments postulated the existence of different "phases" in wool and hair fibers that would today probably be equated

with different protein fractions. From x-ray diffraction studies Astbury later postulated a composite arrangement for wool and hair protein by analogy with muscle proteins. Goddard and Michaelis [17] isolated fractions with different compositions from reduced-wool solutions and Alexander and others [1, 3, 13] have achieved a better separation into more distinct components by using oxidized wool. Rudall [31] has reviewed the evidence from early histological work, and from his own studies on biosynthesis in horn, that a large proportion of the sulfur in the hard keratins, such as wool, is incorporated as a low-molecular-weight sulfur-rich material at or just before keratinization, after the main features of the structure have been established. He also speculated on the possibility that variations in the stability of the hard keratins are due to variations in the amounts of the two fractions. Indirect electron-microscopic evidence for a sulfur-rich matrix in the cortical cells of hair and wool has been reported by Mercer and by Jeffrey et al. [26, 18]. Zahn and Kessler [36] have recently found new chemical evidence for a sulfur-rich cementing material in wool. It is therefore certainly possible that the chief difference between normal and copper-deficient wool is in the proportion of this sulfur-rich material. But it is difficult to prove. The only evidence so far is that copper-deficient wool has fewer disulfide groups, that after oxidation it dissolves less readily in buffer solutions, and that it has a lower proportion of a soluble component than normal.

This raises the question of the nature and origin of the soluble component; experiments on the properties of this material are being undertaken. In Part II amino-acid analyses are described which show that the soluble component differs markedly in composition from the rest of the fiber and that it resembles similar material isolated previously by acid or salt precipitation from high-pH wool protein solutions [12]. In particular, it has twice the concentration of cysteic acid residues as the rest of the fiber. It is also shown in Part II that the insoluble component has the same composition whether it comes from normal or copper-deficient fibers, although the soluble components differ slightly in cysteic acid concentration.

The origin of the soluble component in the fiber is unknown. It is probable that during performic acid oxidation splitting of non-disulfide bonds occurred, but it is most improbable that this oxidation for a few hours at low temperature caused a random hydrolysis of peptide bonds—no such effect has been reported for other proteins—so any peptide hydrolysis or other unwanted splitting would be expected to be confined to particularly labile and therefore atypical bonds. If the soluble component were an artefact produced during oxidation by the splitting of such bonds to give fragments with a high sulfur content and different amino-acid composition, then the above results would still indicate an important difference between the two sorts of wool; if the material split off had originally a structural function, the argument would be unaffected. Incorrect conclusions would, however, be drawn if there were a difference between the two sorts of wool in the distribution of the hypothetical labile bonds. There is no evidence on this, apart from the fact that the tryptic digests of oxidized normal and copper-deficient wool contain a similar mixture of peptides, but it would suggest an even more profound difference between the two sorts of wool than that suggested by taking the evidence on solubility at its face value.

The effect of the bilateral structure of wool fibers on the dissolution experiments has not been discussed, although it has been mentioned that the normal and copper-deficient fibers are alike in this respect, according to measurements on the areas of the two segments after staining cross-sections. Bilateral structure could still be revealed, without apparent change, after the soluble component had been dissolved out of oxidized fibers of both sorts, although it was not possible to tell whether the two segments contained the same or different amounts of the soluble material.

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Part II: The Amino-Acid Composition of Oxidized Normal and Copper-Deficient Wool Fractions and Further Attempts at Fractionation

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Abstract

The amino-acid compositions of oxidized normal and copper-deficient wool from one sheep and of two components separated from them at pH 6 have been measured by ion-exchange chromatography. The differences between the "soluble" and "insoluble" components are those expected from previous work. The "insoluble" components from normal and copper-deficient wool appear to be identical, although there is a small difference in the "soluble" components. Attempts at fractionating the "soluble" components by chromatography, paper electrophoresis, and dialysis gave indications of heterogeneity but were not very successful.

Introduction

In Part I evidence was presented suggesting that in oxidized normal and copper-deficient merino wool there are different proportions of two protein components, the smaller of which is soluble at pH 6 and the larger insoluble at this pH. As a further step in this investigation into the reasons for the abnormal physical behavior of copper-deficient wool, amino-acid analyses have been done on these components from wool of each sort and on the oxidized wool before fractionation. As expected from previous work [7], the soluble and insoluble components were found to have different amino-acid compositions, the chief difference being in the concentrations of cysteic acid, aspartic acid, proline, leucine, and lysine. The composition of the insoluble component was found to be the same whether it was derived from normal or copper-deficient wool, but the soluble components differed slightly in cysteic-acid concentration. This led to attempts at the fractionation of the soluble component, some of which are described here.

Materials and Methods

Wool Samples

The normal and copper-deficient wool samples used for analysis and for fractionation experiments were from sheep E5, the growth of which has already been described [5].

Amino-Acid Analysis

The original ion-exchange chromatographic method of Moore and Stein [11] was used, with modifications suggested by later papers and by experience in this laboratory [3, 4]. In brief: the acidic and neutral amino acids were separated on a 100-cm. column of pulverized Amberlite IR 120 (200–400 mesh). During elution the buffer was changed from pH 3.10 to 5.0 and the operating temperature from 30° to 50° and then to 70°. To improve the separation of serine and threonine, the column was sometimes eluted with a buffer of pH 3.40 until after the emergence of aspartic acid, when the pH 3.10 buffer was used. The basic amino acids were separated as usual on a 15-cm. column of the same resin.

The amino-acid concentrations were measured by the ninhydrin method of Moore and Stein [12], slightly modified by the use of a stronger aqueous buffer [4].

For analysis of the whole oxidized wool, the sample (0.100 g., of known moisture content) was oxidized for 4.5 hr. at 5° with 2.5 ml. of performic acid as described previously [5], although a glass tube was used instead of a flask. After the acid had been sucked off, the sample was left over sodium hydroxide under vacuum for 6 days or longer before the tube was sealed up with 20 ml. of twice-distilled constant-boiling hydrochloric acid. The mixture was then heated for 24 hr. at 103°, and after the acid had been evaporated off it was made up to standard volume with buffer solution. Portions of 2.0 or 3.0 ml. were applied to the columns.

The soluble and insoluble components of oxidized wool were separated by shaking the oxidized sample (0.100 g.) with pH 6 pyridine-acetate buffer (20 ml.) for 7 hr. at 30° as before [5]. The insoluble component was filtered off, dried over phosphorus pentoxide, and weighed before acid hydrolysis, as for the whole wool. The filtrate was freeze-dried and the soluble component in it was weighed after being dried over phosphorus pentoxide.

Two chromatographic columns were used; the normal and copper-deficient wools or wool fractions were analyzed at the same time to facilitate comparison of the results.

Results

Amino-Acid Analyses

Table I gives the amino-acid composition of the soluble and insoluble components of normal and copper-deficient wool E5 and of the oxidized wools before fractionation, expressed first as grams of amino acid from 100 g. of dry oxidized protein and then as μmoles/g. of dry *unoxidized* protein. The figures for unoxidized wool are useful for comparison with other results but required the application of correction factors, discussed below, for the increase in weight on oxidation.

The results in Table I are averages of two or more estimations, and the errors were apparently similar to those already reported for other amino-acid analyses in this laboratory [4]. As before, our results for arginine are unreliable. No corrections have been applied for losses during hydrolysis and chromatography or for incomplete hydrolysis. This should not affect the comparisons which were the

TABLE I. Amino-Acid Compositions: Wool E5 Normal and Copper-Deficient, Whole Wool and Fractions after Oxidation

Amino acid	Soluble component				Insoluble component				Whole wool			
	Normal		Cu-deficient		Normal		Cu-deficient		Normal		Cu-deficient	
	g./100	μmol./g.	g./100	μmol./g.	g./100	μmol./g.	g./100	μmol./g.	g./100	μmol./g.	g./100	μmol./g.
$CySO_3$	27.8	1,860	22.8	1,500	13.6	865	13.7	870	17.8	1,140	15.5	995
Asp	4.4	360	4.5	365	7.9	620	8.4	660	7.6	610	8.0	640
Thr	9.0	830	9.0	820	5.7	505	5.8	510	6.3	560	6.7	595
Ser	9.4	985	9.5	980	9.3	930	9.3	930	8.5	860	8.8	890
Glu	10.3	770	10.6	780	14.2	1,010	14.5	1,015	13.5	980	13.3	960
Pro	14.9	1,430	13.9	1,300	4.8	440	4.9	450	8.2	760	7.7	710
Gly	5.5	810	5.4	780	6.3	880	6.4	900	5.6	795	5.9	745
Ala	2.6	320	3.0	365	4.5	530	4.3	510	3.7	440	3.5	415
Val	6.6	620	6.4	590	5.7	510	5.6	500	6.5	590	6.3	570
Leu	5.2	435	5.2	430	8.9	715	9.1	730	8.2	665	8.0	650
Ileu	3.0	250	2.9	240	3.4	270	3.6	290	3.9	310	3.9	320
Tyr	3.5	210	3.5	210	2.7	160	3.0	175	3.1	180	3.1	180
Phe	4.0	270	4.1	270	3.4	215	3.6	230	3.7	240	3.5	225
His	1.5	105	1.8	125	1.5	100	1.3	90	1.3	90	1.0	70
Lys	2.7	220	2.8	210	4.4	315	4.2	300	3.0	220	3.2	230
Arg	10.0	630	7.6	470			7.6	460	8.4	515	7.7	465

chief object of this work. Methionine sulfone was not detected. It would be expected in small amount only, and it probably emerged from the 100-cm. column with serine [3]. From the low results for tyrosine, the presence of chlorotyrosine was suspected [3, 15]. An unidentified peak which may have represented this was found between those of ammonia and arginine, but it was not measured because a standard was not available.

Table I shows that the soluble and insoluble components are very different in composition, the soluble component having more cysteic acid, threonine, and proline but less aspartic acid, alanine, leucine, and lysine. The soluble component is similar in composition to the sulfur-rich fraction analyzed by Corfield, Robson, and Skinner [7] (so-called "γ-keratose"), although a quantitative comparison reveals differences which would be partly but not entirely explained if our material contained more of other fractions of the fiber. This would probably be expected from the method of isolation, and some evidence for it is given in the next section. Table I also shows that there is a greater difference in the cysteic acid concentration of the two components than that given in a preliminary report [6].

According to Table I, the insoluble components of normal and copper-deficient wool have the same composition, and this is true of the soluble components with the exception that the soluble component from normal wool has significantly more cysteic acid.

It was found that the cysteic acid analyses on oxidized wool by the method of Moore and Stein gave results in good agreement with those from cystine analyses on unoxidized wool by the phospho-18-tungstic-acid method [13]. Table II compares cysteic acid and half-cystine plus cysteine concentrations in normal and copper-deficient wool from sheep E5. Two figures are given in Table II for cysteic-acid concentration; the first was measured directly on the whole wool and the second calculated from the analysis of the soluble and insoluble components separated from it. Results for most of the other amino acids also agree reasonably well when compared in this way, although serine, *iso*leucine, and arginine show inexplicable divergences.

In calculating the composition of the soluble and insoluble components in terms of the weight of unoxidized protein, it was necessary to apply a correction for the increase in weight on oxidation. This was also necessary in calculating the proportions of the two components from the oxidized weight of one of them—this is discussed in Part I. On oxidizing samples of wool E5 for 4.5 hr. in the usual way in performic acid and then drying them to constant weight at 103°, the average increase in weight was found to be 6.4% for normal wool and 5.9% for copper-deficient wool. The increase in weight expected from the conversion of all the cystine to cysteic acid was 5.2% for normal and 4.6% for copper-deficient wool. The excess of the experi-

mental over theoretical values, about 25%, probably represented the oxidation of minor constituents, such as methionine, tryptophane, and possibly tyrosine. No measurable residue was left on evaporation of the performic acid used for oxidation. The increase in weight of the soluble and insoluble components due to oxidation was calculated from the cysteic acid percentages in Table I as 3.9% for the insoluble components, 8.1% for the normal soluble component, and 6.6% for the copper-deficient soluble component. Assumption that the increase due to other factors represents 25% of these, as it does for whole wool, gives a 5.0% increase for the insoluble components, a 10.1% increase for the normal soluble component, and an 8.2% increase for the copper-deficient soluble component. These figures were used to obtain the unoxidized weights of the two components. Adding up the unoxidized weights of the components isolated from a known weight of wool gave good agreement with the original total weight, so that the corrections were probably satisfactory. This use of the correction factors is illustrated in Table III, which also provides another example of the different proportions of the two components in normal and copper-deficient wool.

Apart from the difference in cysteic acid concentration, the results in Table I reveal no significant differences between oxidized normal and copper-deficient wool, in agreement with previous reports on the analysis of unoxidized wools [10]. According to the postulate that the chief difference between normal and copper-deficient wool is in the proportion of the soluble fraction, the copper-deficient wool would be expected to have an excess of those amino acids that are present in greater amount in the soluble component. Except for cysteic acid, the differences expected are very small. A difference of 7% would be expected for proline, and although the difference in proline concentrations in Table I is of about this magnitude, it is not significant because we had difficulty in estimating this amino acid. Very accurate analyses of proline, leucine, lysine, and aspartic acid would help to confirm or refute the postulate.

The results for normal wool in Table I show that, apart from those of tyrosine and arginine, the amino-acid concentrations agree reasonably well with previous analyses on merino wool. Tyrosine was presumably lost as chlorotyrosine during acid hydrolysis [3]. There was no loss of lysine, such as reported after prolonged oxidation in aqueous peracetic acid [8].

Preliminary Attempts at Fractionating the Soluble Component

Because the analyses reported above suggested a slight difference in the amino-acid composition between the soluble components from normal and copper-deficient wool, attempts were made at fractionating these components. It was first found, by using the procedure described in Part I, that about 12% of the soluble component precipitated on acidifying the solution to pH 3.5, although it was difficult to collect and weigh the small precipitates accurately. Presumably a constant proportion of the "insoluble" component went into solution and then precipitated in acid.

The behavior of the soluble component during dialysis was examined as follows: a standard solution of this component containing about 1 mg. in 5 ml. of water or buffer was dialyzed in a cellophane tube against running tap water. The dialyzed solution was then made up to 10 ml. and the amount of protein that did not penetrate the membrane was estimated by the Folin-biuret method [5, 9]. With freshly isolated material, reproducible results were obtained which showed that a negligibly small

TABLE II. Comparison of Results for the Analysis of Cysteic Acid and Cystine (Whole Wool E5)

	Amino-acid concentration, μmol./g. of unoxidized wool	
	Normal	Cu-deficient
$CySO_3$ (direct measurement)	1,140	995
$CySO_3$ (calculated from components)	1,115	1,000
CyS-* + CySH	1,076	964

* Half cystine.

TABLE III. Application of Correction Factors

	Weight, g.	
	Normal	Cu-deficient
Dry unoxidized whole wool	0.0988	0.0971
Oxidized soluble component	0.0312	0.0239
Unoxidized soluble component	0.0281	0.0221
Oxidized insoluble component	0.0750	0.0811
Unoxidized insoluble component	0.0714	0.0772
Unoxidized soluble + insoluble components	0.0995	0.0993

amount of protein passed through the membrane in 24 hr. and that this was not increased by dialysis for another 24 hr. There was no difference between the normal and copper-deficient soluble components. With some early preparations, however, erratic results were obtained, and a large proportion of the protein passed through the membrane. The reasons for this have not been investigated, but the samples were not freshly isolated, and it is possible that a protein with such a high concentration of cysteic-acid residues would readily exhibit the effects of autohydrolysis.

Attempts were made at chromatographing the soluble component using ion-exchange resins. Very little of this component was retained by acidic resins in the hydrogen form, but it was held strongly by basic resins; it was almost impossible to elute it from these. Up to 70% of the soluble component could, however, be eluted from diethylaminoethyl cellulose (DEAE cellulose) [14], and many tests were done on columns of this material. Figure 1 shows a typical curve obtained by chromatographing the soluble component of normal wool E5 (3 mg.) on a column 17 cm. long and 0.6 cm. in diameter, eluting with sodium acetate solutions. The fractions (1.0 ml.) were analyzed by the Folin-biuret method. From the shapes of the curves, most of the fractions appear to be complex mixtures. The number and positions of the peaks were reproducible, but their sizes were not. The soluble component from copper-deficient wool gave qualitatively similar curves to those from that of normal wool, but because of the lack of reproducibility it was not possible to tell whether it contained the same or different proportions of the various fractions or, indeed, whether the different peaks represent different protein fractions. Further experiments on this subject are being undertaken.

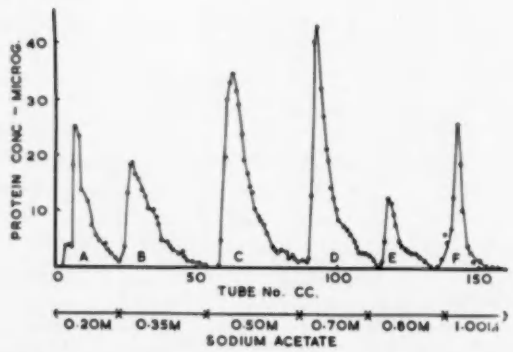

Fig. 1. Chromatography of 3 mg. of the soluble component from oxidized wool E5 (normal) on a DEAE cellulose column.

Attempts at separating the soluble component by paper electrophoresis were unsuccessful. Much of this component moved towards the anode, but there were no discrete bands and it was difficult to detect the component on paper.

It might be possible to effect a fractionation of the soluble component by taking advantage of the fact that some of this material dissolves in the formic acid during oxidation (as first observed by Blackburn [2]) although this was not investigated.

Discussion

The possible importance of the evidence for different proportions of "soluble" and "insoluble" components in normal and copper-deficient wool has already been discussed in Part I. The amino-acid analyses reported here show that the method of separation used gives components with different amino-acid compositions that resemble those isolated previously by precipitation methods, and that, with the exception of a lower proportion of cysteic acid in the soluble component of oxidized copper-deficient wool, the composition of the components is independent of the sort of wool.

Attempts were made to fractionate the soluble components in case the difference between the two sorts of wool is confined to one fraction of these. The behavior of the soluble components on chromatographic columns, however, was erratic; it was impossible to get reproducible results. This suggests that a thorough study of the physical properties of the soluble component is desirable, and such a study has been planned.

The dialysis experiments suggest that the soluble component contains little material with molecular weight less than about 20,000. It is therefore possibly less degraded than the "γ-keratose" fractions described previously, for which an average molecular weight of 4,500 has been reported [1].

Acknowledgment

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report is published by permission of the Council for Scientific and Industrial Research.

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Variations in the Equilibrium Weight of Wool at Zero Humidity

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Abstract

The equilibrium weight of a wool sample obtained on desorption *in vacuo* is dependent on the previous treatment of the sample with respect to water. Exposure to $< 5\%$ RH has no effect on the subsequent equilibrium weight *in vacuo*, but exposure to intermediate relative humidities causes an increased equilibrium weight *in vacuo* on subsequent desorption. The maximum rate of increase occurs on exposure to approximately 50% RH. Exposure to high relative humidities (near 100%) does not cause an increase in the subsequent equilibrium weight *in vacuo*.

Initially the increase of equilibrium weight *in vacuo* is linearly dependent on the time of exposure to a particular relative humidity. Increases are less marked at higher temperatures ($> 65°$ C.). Increases of the equilibrium weight *in vacuo* can be removed by saturation with liquid water or water vapor prior to desorption.

Introduction

In a study of the vacuum dry weight of a wool sample by Watt, Kennett, and James [3], it was reported that the equilibrium weight obtained for a particular wool sample was dependent on the conditions of desorption. In addition, it was noted that the equilibrium weight of a wool sample at zero humidity, obtained after a standard desorption procedure, varied according to the previous history of the sample with respect to its contact with water vapor.

Evidence will be presented in this paper showing that the initial water concentration, the temperature, and the length of time wool fibers were equilibrated with water vapor prior to desorption are some of the factors influencing the equilibrium weight obtained after vacuum drying. Changes of up to 3% in the equilibrium weight have been observed, and these changes are not affected by continued evacuation.

A reproducible equilibrium weight can be obtained on desorption to zero humidity by ensuring that the wool sample is saturated with water for some time

prior to the desorption. Thus, even if the equilibrium weight obtained by vacuum drying of a wool sample has been altered by the methods outlined in this paper, it is possible to return to the original value, showing that there has been no permanent damage to the wool fibers as indicated by weight changes.

Experimental Method

The weights of the wool samples were determined by suspending them from calibrated quartz helical springs in sorption chambers and following the changes of spring length with cathetometers. Weight changes of 0.005% could be detected. One apparatus which has been described previously [3] could be immersed in a liquid thermostat and a second apparatus described by Watt [4] was housed in an air thermostat. Load–extension curves were obtained on a Cambridge Textile Extensometer, the fiber being mounted in the form of a loop, as described by Feughelman [1].

The wool samples were taken from a pen-grown Merino fleece or from a Corriedale fleece. They were cleaned by several washings with cold petroleum ether followed by thorough washing with distilled water until the pH of the rinse water was unaltered by contact with the wool.

The concentration of water in the fibers is termed the regain and is the weight of water expressed as a percentage of the equilibrium weight *in vacuo* at that temperature; the concentration of water vapor surrounding the fibers is expressed as the relative humidity (RH).

Results

The Effect of Relative Humidity

In the course of kinetic measurements of the uptake of water vapor by wool it became apparent that wool samples which had been exposed to an intermediate relative humidity (e.g., 50%) returned to a higher equilibrium weight than that previously obtained on desorption to zero humidity at the same temperature. On the other hand, wool samples which had been saturated with water vapor showed no change in the equilibrium weight *in vacuo*.

In order to correlate the vacuum equilibrium weight with the humidity prior to desorption, a Merino wool sample was saturated and desorbed at 45° C. to give the initial equilibrium weight *in vacuo*. The sample was then exposed to successively increasing humidities for a standard time of 17 hr. Between each exposure the sorption chamber was evacuated for 7 hr. to give the equilibrium weight *in vacuo*. The temperature was maintained at 45° C. The values of the relative humidity to which the wool sample was exposed and the equilibrium weight of the sample after evacuating the chamber are given in Table I.

The fact that no increase in the equilibrium weight *in vacuo* was observed after exposure to 4.5% RH is in agreement with experiments in which Corriedale wool at 35° C. was exposed to water vapor of relative humidity up to 5.0% over a period of 8 days and Corriedale wool at 50° C. was exposed to relative humidities up to 5.0% for 21 days; no increase in the equilibrium weight on subsequent desorption to zero humidity was observed. However, exposure

TABLE I

Number of exposure	Relative humidity, %	Equilibrium weight *in vacuo*, mg.	Change of equilibrium weight, %
1	4.5	40.704	—
2	25.1	40.781	0.19
3	45.7	40.867	0.40
4	63.1	40.924	0.54
5	78.0	40.989	0.70
6	90.4	41.005	0.74
7	100.0	40.908	0.50
8	100.0 for 2 days	40.704	—

TABLE II

Time of exposure at 50% RH, hr.	Percentage increase of equilibrium weight *in vacuo*
24	0.21
48	0.39
72	0.59
96	0.75
288	1.87

TABLE III

Temperature, ° C.	Time of equilibration with 50% RH, hr.	Percentage increase of equilibrium weight *in vacuo*
19.0	283	2.00
35.0	288	1.99
45.0	288	1.87
50.0	288	1.73
64.7	288	1.71
80.0	288	0.90
100.0	288	0.22

to relative humidities above 5% invariably caused an increase of the equilibrium weight *in vacuo*.

After the fiber was exposed to a relative humidity of 25.1% the equilibrium weight *in vacuo* showed a marked increase and increased further with each desorption after exposures up to 90% RH. The equilibrium weight *in vacuo* decreased after the wool sample had been saturated with water vapor; it was found that, after the fiber was saturated with water vapor for a total of three days, the equilibrium weight *in vacuo* had decreased to its initial value. Similar behavior was observed with a sample of Corriedale wool.

The Effect of Time of Contact with Water Vapor

To determine the effect on the final equilibrium weight of the length of time for which the wool sample was equilibrated with water vapor prior to desorption, a wool sample which had been desorbed from saturation *in vacuo* was exposed to 50% RH at 45° C. for the standard period of 17 hr. and subsequently desorbed *in vacuo*. This cycle was repeated; in Figure 1 the increase in equilibrium weight *in vacuo* is plotted against the total number of hours at 50% RH.

Other wool samples which had been saturated and desorbed at 45° C. were exposed to a relative humidity of 50% for varying lengths of time prior to desorption. As can be seen from Table II, the increase in the equilibrium weight of the wool sample is initially almost proportional to the length of time that the wool was exposed to water vapor prior to desorption. The results of Figure 1 and Table II taken together show that the cycling action of absorption–desorption does not contribute to the increase of equilibrium weight. Additional experiments also showed that the rate of absorption or desorption did not alter the magnitude of the change of equilibrium weight *in vacuo*.

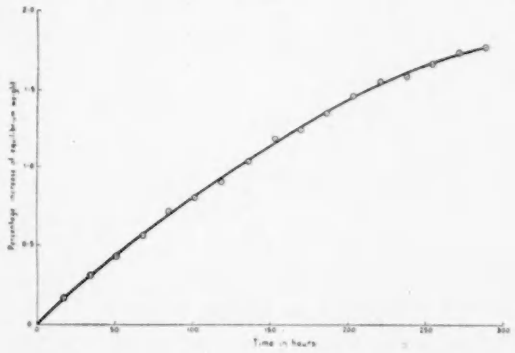

Fig. 1. Percentage increase of equilibrium weight *in vacuo* as a function of the time of exposure to 50% RH, temperature 45° C.

The Effect of Temperature

The increase of the equilibrium weight *in vacuo* of Corriedale wool samples after exposure to a relative humidity of 50% for a period of 12 days and subsequent desorbing was determined at a number of temperatures. The results are given in Table III. It is apparent that the effect of temperature is slight up to 64.7° C., but the increase of the equilibrium weight *in vacuo* appears to be more readily achieved at lower temperatures.

The increase in the equilibrium weight *in vacuo* achieved at 80° C. is much less than the weight increases observed at lower temperatures. Cooling to 20° C., saturating with water vapor, and subsequent desorption returned the wool sample to its original equilibrium weight. After exposure at 100° C. for 12 days to a relative humidity of 50%, a wool sample appeared slightly yellow and on subsequent desorption the equilibrium weight *in vacuo* was 0.21% less than the original weight. Cooling to 20° C., saturation with water vapor, and subsequent desorption caused a further decrease of 0.22% in the equilibrium weight.

The results obtained at 100° C. have been interpreted as an increase of the equilibrium weight *in vacuo* of 0.22% as a result of the exposure of the wool sample to 50% RH and a simultaneous decrease of 0.43% in the weight of the sample due to degradation. The fact that a wool sample can be returned to its original equilibrium weight after exposure to 50% RH at 80° C. shows that no degradation, as measured by loss of weight, occurs under these conditions.

The effect of temperature on the equilibrium weight *in vacuo* was determined by heating two Merino wool samples with continuous evacuation from 20° C. Both samples were desorbed from saturation at 20° C. to obtain the initial equilibrium weight *in vacuo*, and one sample (Sample B) was given an increase of 0.87% in the equilibrium weight *in vacuo* while Sample A was maintained at the initial weight. The two samples were heated with constant evacuation. The temperature was held constant at various intervals for a length of time equiva-

lent to the time required to desorb from saturation at that temperature (or for 10 min. at temperatures above 100° C.). The loss of weight as a function of temperature is illustrated by Figure 2. It can be seen that the loss of weight with increasing temperature is identical in the two samples up to 80° C. Above 80° C. Sample B clearly loses more weight than Sample A and at 140° C. has lost an additional 0.04% of the initial equilibrium weight *in vacuo* at 20° C. Maintaining the temperature at 140° C. for a further half-hour caused a weight loss of 0.10% to Sample B. An additional half-hour period at 140° C. failed to change the weight of either sample.

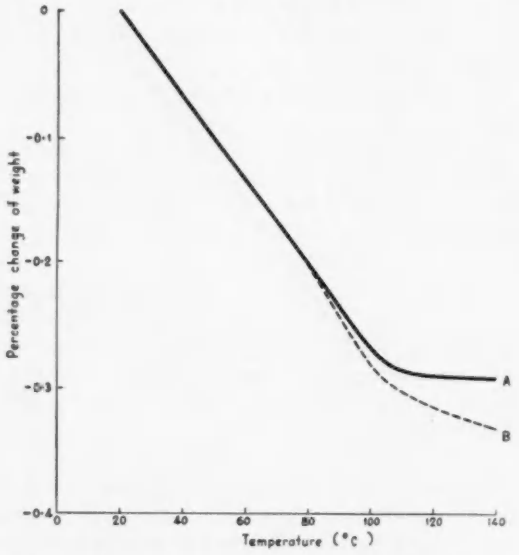

Fig. 2. Change of weight vs. temperature taking the equilibrium weight *in vacuo* at 20° C. as zero. —— Wool which had been dried from saturation at 20° C. - - - - - - Wool which had been given an increase of 0.87% in the equilibrium weight *in vacuo*.

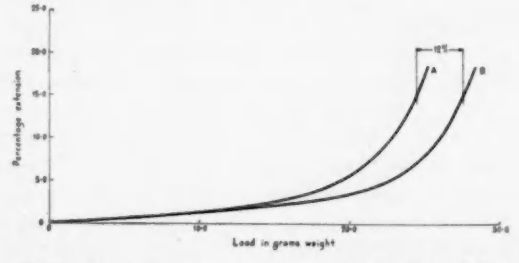

Fig. 3. Load vs. extension curves at 17% RH. (B) A Corriedale fiber with an increase of 2% in the equilibrium weight *in vacuo;* (A) the same fiber after immersion in distilled water at 52° C. for 1 hr.

Cooling of the wool samples to 20° C. followed by reheating to 140° C. did not change the weight of Sample A but further decreased the weight of Sample B. Two such temperature cycles caused a weight loss of 0.14% in Sample B, which was then only 0.59% heavier than Sample A instead of the 0.87% before heating. Further temperature cycling was not effective in reducing the weight of Sample B.

The wool samples were not heated above 140° C., as it has been previously reported by Watt, Kennett, and James [3] that at this temperature the weight becomes constant with increasing temperature and at higher temperatures thermal degradation of dry wool occurs.

The Effect of Equilibrium Weight on Mechanical Behavior

A Corriedale wool sample was placed in water at 52° C. for 1 hr. prior to desorption *in vacuo* to the equilibrium weight at 19° C. The sample was equilibrated with water vapor at 50% RH for 283 hr. and then desorbed to a new equilibrium weight *in vacuo* which was 2% higher than the former. A portion of the sample was replaced in water at 52° C. for 1 hr., and on desorption it was found that the increase in equilibrium weight had disappeared.

A wool fiber from the remaining portion of the sample was placed in a Cambridge extensometer in a dry atmosphere of air and then equilibrated with 17% RH in air. A load–extension curve was taken with a rate of loading of 4 g./mm. up to 17.5% extension and is shown as Curve B of Figure 3. An extension of 17.5% was chosen, as this value is well into the yield region for Corriedale fibers and the extension is linear as a function of load. Corriedale fibers are not permanently damaged at this extension. The load was taken off and the fiber placed in distilled water at 52° C. for 1 hr., desorbed in a dry atmosphere of air, and again equilibrated with 17% RH in air. The load–extension curve was repeated (Curve A of Figure 3). It can be seen that in order to extend the fiber by 15% the load required was 12% greater before the fiber was relaxed in water. Six fibers of this sample and three fibers from another sample with a similar weight increase were tested in this manner with the same result.

In order to test whether immersion in water at 52° C. would bring about a change in the mechanical behavior of wool irrespective of the increase in equi-

librium weight *in vacuo*, a fiber was immersed for 15 min. prior to the above series of experiments. The curves obtained before and after the fiber was placed in water at 52° C. for 1 hr. were then the same and identical with Curve A of Figure 3.

The load–extension measurements were made after absorption at 17% RH because it was found that dry fibers tend to break before 15% extension and because water content equilibrium can be more quickly established on absorption than desorption.

Discussion

An increase in the equilibrium weight *in vacuo* of a wool sample can be achieved by allowing that sample to remain in contact with water vapor at an intermediate relative humidity. The weight can be decreased to the original equilibrium weight by saturating the wool sample with water vapor before evacuating. These facts suggest that the wool fibers have not undergone any permanent change and that the change in weight of the wool sample *in vacuo* represents a change of water content. The additional water molecules retained in the wool fibers may be bound with high energy or they may be trapped inside the wool fibers; i.e., water of occlusion.

Saturation and subsequent desorption does not bring about any increase in the equilibrium weight *in vacuo*. Therefore it is unlikely that the increase in weight *in vacuo* can be attributed to water penetrating the fibers in the swollen state and then being unable to pass through the dry outer layers of the fiber on desorption, as on saturation the fibers would be in their most swollen condition. If the increase of the equilibrium weight *in vacuo* is due to occlusion of water it is more probable that the configuration of the molecular chains in the fibers does not permit the escape of the water molecules.

On the other hand, if exposure to an intermediate relative humidity causes a number of high-energy sites to be made available to water molecules, these sites would need to be of higher energy than the hydrophilic side chains which exist in wool, as it has been shown by Watt, Kennett, and James that the high-energy NH_3^+ water bonds are broken on vacuum desorption. The energy of bonding between the water molecules and the wool would need to be sufficiently high to have an equilibrium vapor pressure of less than 10^{-3} mm. mercury, as the forms of desorption curves were found to be the same irrespective of the final equilibrium weight *in vacuo*.

The time required to establish weight equilibrium at 45° C. for integral absorption at 50% relative humidity by Corriedale fibers is approximately 30 min. Consequently the figures of Table II clearly show that the causes of the change in equilibrium weights *in vacuo* are occurring long after equilibrium has been established in the fibers with respect to water vapor uptake. That is, while the fibers are exposed to an intermediate relative humidity they are assuming a form which enables them to retain more water on desorption without absorbing additional water in the process. On the other hand, the initial weight *in vacuo* is obtained by desorption from saturation, and subsequent saturation of the wool fibers yields the same weight *in vacuo*, indicating that the configuration of the molecular chains is the same after each saturation.

The tests on the mechanical properties of fibers with an increased equilibrium weight *in vacuo* showed that these fibers were stiffer than the fibers which had been desorbed from saturation; i.e., more load was required to extend the fibers to the same extent. It has been shown by Speakman [2] that an increased relative humidity surrounding a fiber brings about a decrease in its stiffness. Therefore, in this case, where the fibers containing more water showed increased stiffness, it is apparent that the structure of the fibers has undergone a change of molecular organization.

It is not possible at this stage to decide whether the additional water is held with high energy or occluded due to the structural changes in the fibers. It could be argued on the one hand that the water molecules became occluded during the configuration changes of the fibers and that the increased thermal agitation at higher temperatures allows some of these molecules to escape. On the other hand, it may be postulated that exposure to an intermediate relative humidity, which leads to the breaking of some interchain bonds, allows the slow buildup of cooperative bonds from various molecular chains to the same water molecule, whereas in the fully swollen fiber these molecular chains would be too far apart. These bonds would be less stable at high temperatures and would permit the escape of some of the water molecules.

It is well known that swelling of polymeric materials is accompanied by the breaking of secondary interchain bonds. In the wool–water system the maximum swelling is produced by saturation of

the wool fibers, and it is probable that this swelling causes the rupture of many of the weak interchain bonds. The fibers would assume a preferred orientation in their most relaxed position. It has been reported by Watt [4] that absorption to high humidities by wool fibers shows large overshoots of the water vapor uptake. The overshoots can be attributed to the breaking of many weak interchain bonds at high water contents followed by the re-formation of some of these bonds, not necessarily in their original positions. Presumably similar behavior occurs on absorption to intermediate relative humidities (e.g., 50%), although to a smaller extent and more slowly, so that uptake overshoots are not observed. It is probable that the wool fibers assume a more ordered configuration as a result of the reformation of interchain bonds broken on swelling. This indicates that, on absorption of water vapor at intermediate relative humidities, wool fibers first reach an uptake equilibrium followed by a long term change towards a structural equilibrium.

Exposure to water vapor at low relative humidities does not bring about any change in the equilibrium weight *in vacuo* or any change in the mechanical properties of the fibers. It is probable that absorption at such low relative humidities ($< 5\%$ RH) does not swell the fibers sufficiently to alter the position of the molecular chains relative to each other.

It was noted that, for any one wool sample which had been taken through the cycle of an increased equilibrium weight *in vacuo* followed by a return to the original equilibrium weight, subsequent increases in the equilibrium weight *in vacuo* were more readily attained. Thus, although the results in this paper are quantitative when comparative experiments have been made, in cases where the wool has been taken from different samples the differences of pretreatment may be responsible for differences in the magnitude of the change of equilibrium weight *in vacuo* under a particular set of conditions.

This paper has dealt exclusively with the wool–water vapor system in the absence of air. A few experiments performed with air present in the sorption chamber before admitting water vapor showed similar variations of the equilibrium weight on subsequent desorption, but the magnitude of the equilibrium weight changes was approximately one-half the size of the changes observed in the absence of air.

Although the largest change of equilibrium weight *in vacuo* reported in this paper is 2.0% after 283 hr. exposure to 50% relative humidity at 19° C., larger changes can be obtained. We have observed an increase of 3.4% in the equilibrium weight *in vacuo* with a wool sample which was mounted in the sorption chamber for 75 days and subjected to a variety of sorption steps at various temperatures. The whole of the increase was subsequently removed by saturation and desorption.

Acknowledgment

The authors wish to thank Mr. M. Feughelman for performing the load–extension experiments described in this paper.

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Effects of Cross-Linkage in Wrinkle-Resistant Cotton Fabrics

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Abstract

The cross-linking of cellulose in cotton fabric with three wrinkle-resistance finishing agents has been investigated. The three agents used were dimethylol urea, dimethylol ethyleneurea, and formaldehyde. With all three compounds, maximum crease recovery angles are obtained with a substitution of 0.04–0.05 cross links per anhydroglucose unit. As crease recovery angles increase, the elongation and permanent set caused by an 8-kg. tensile load on a 1-in. fabric strip decreases. Permanent set approaches zero near 135° warp crease recovery. Changes in the load–elongation curves indicate elastic modulus increases as crease recovery angles increase. Treatment of fabric to the same crease recovery angles with the three compounds reduces tearing and breaking strengths to different extents. The differences are due to different amounts of cellulose degradation caused by the acidic catalyst systems and not to the structure of the cross links.

Introduction

The treatment of cotton fabrics to impart wrinkle resistance is now a commonplace finishing operation. In addition, most of the presently used wash-and-wear finishing treatments are essentially only extensions of the wrinkle-resistance treatments. These treatments usually consist of the application to the fabric of methylol derivatives of ureas or other organic amides with acidic catalyst. The production of wrinkle resistance by such methods has been attributed, although possibly with less than conclusive evidence, to the formation of cross links between the polymeric molecules of cellulose [3, 5, 7]. Presumably this cross-linking reaction occurs only with the cellulose in the accessible regions of the cotton fiber.

Cross-linking of cellulose in cotton fabrics has been shown to produce the desired wrinkle resistance by changing the elasticity of the cotton fiber and giving the fabric the ability to recover from wrinkling or creasing [2]. The wrinkle resistance is always accompanied, at least at the present time, by losses in strength and extensibility. In this report are described the properties imparted to cotton fabric by three cross-linking agents of known effectiveness in producing wrinkle resistance. An attempt is made to correlate changes in the various fabric properties with each other, with the degree of cross-linking introduced, and with the structure of the cross link formed.

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Materials and Methods

Three series of treated cotton fabric were prepared by the application of three cross-linking agents over a range of add-ons. The three agents used were dimethylol ethyleneurea (DMEU), dimethylol urea (DMU), and formaldehyde (CH_2O). The two dimethylol compounds are typical of the type of agent in commercial use, while formaldehyde serves as an example of a cross-linking agent of smaller than usual molecular size. Dimethylol urea, although often considered a resin-former rather than cross-linking agent, has been shown [5] to form cross links when applied to cellulose.

The cross-linking agents were laboratory preparations. The dimethylol compounds were prepared by the treatment of the urea with two molar equivalents of formaldehyde in a slightly alkaline solution. These solutions, of 20% concentration for dimethylol urea and of 33% concentration for dimethylol ethyleneurea, were diluted and used after standing overnight. A 20% formaldehyde solution was prepared by dissolving paraformaldehyde in water under reflux.

Zinc nitrate was used as catalyst with all three agents. Acetic acid, 10% of the weight of zinc nitrate hexahydrate, was added with the dimethylol amides to neutralize any residual alkalinity in the agent. The amount of catalyst used in each applica-

tion was adjusted so that, as far as possible, each sample in a series treated with one of three agents would undergo reaction at the same acidity. For the series of samples treated with formaldehyde, all the treating baths were made to 0.5% zinc nitrate hexahydrate on the assumption that variations in formaldehyde concentrations made no difference in acidity under reaction conditions. The nitrogenous compounds, however, have been shown to influence the acidity of zinc nitrate in these treatments [3]. Therefore, the methylol amides were applied with the same ratio of catalyst to cross-linking agent in each application. This gives a constant ratio of zinc nitrate to methylol amide, or acid to base, throughout each of the two series, although the concentration of the resulting salt or complex does vary. For dimethylol ethyleneurea and dimethylol urea, the amounts of zinc hexahydrate and acetic acid used were 7% and 0.7% respectively of the dimethylol amide. For each of these two series, a catalyst control was run which was treated with a solution containing 5.8% ethyleneurea or 5.0% urea, the amide concentrations equivalent to 10% of the dimethylol compounds, along with 0.7% zinc nitrate hexahydrate and 0.07% acetic acid, the amounts used with 10% solutions of the dimethylol compounds. For the formaldehyde series, the catalyst control was treated with 0.5% zinc nitrate hexahydrate only.

The fabric used in these investigations was an 80 × 80 cotton print cloth which had been desized, caustic-boiled, and bleached. Samples of this fabric were treated with solutions of various concentrations of the three cross-linking agents to make the three series with varying add-ons and degrees of cross-linkage. The solutions were applied with squeeze rolls, using two dips and two nips and a roll pressure to give 75–85% wet add-on. Drying and curing were performed with the samples held to original dimensions on horizontal pin frames in ovens with circulating air. Samples treated with dimethylol ethyleneurea or formaldehyde were dried at 60° C. for 7 min. and cured at 160° C. for 3 min. The samples treated with dimethylol urea were dried at 60° C. for 10 min. and cured at 150° C. for 3 min. After being cured, all samples were washed with an alkaline detergent solution and tumble-dried. The blank fabrics were washed and dried similarly.

Fabric samples were tested for wrinkle resistance by the Monsanto crease recovery method and for tearing strength by the Elmendorf method, using procedures described by ASTM [1]. Wet crease recovery was also determined by the Monsanto method using samples that had been soaked in detergent solution and blotted, but not dried, before testing. Breaking strengths and elongations were determined on an Instron Tensile Tester, using a 5-in. gauge length and a cross-head speed of 0.5 in./min. Unless otherwise specified, all tests were run at 70° F. and 65% relative humidity on samples conditioned in this atmosphere.

The nitrogen content of the fabric samples was determined by the Kjeldahl method. Formaldehyde residues on the samples were determined by digesting a weighed sample of fabric in 200 ml. of 5% sulfuric acid containing 10 g. sodium sulfate and simultaneously distilling until 160 ml. of distillate were collected. The formaldehyde in the distillate was determined by the method of Yoe and Reid [8]. Analytical values were determined and are reported on the basis of the dry sample. Nitrogen and form-

TABLE I. Properties of Fabrics Treated with Dimethylol Ethyleneurea

Treating solution		Fabric analysis		Fabric properties							
				Crease recovery angle, degrees		Wet crease rec. angle, degrees		Tearing strength, g.		Breaking strength, kg., W	Elongation at break, %, W
% DMEU	% $Zn(NO_3)_2 \cdot 6H_2O$*	% N	% CH_2O	W	F	W	F	W	F		
0	0.00	—	—	89	91	85	104	1040	953	22.1	10.6
0†	0.70	—	—	91	96	80	78	980	693	19.8	10.5
1	0.07	0.11	0.16	98	88	85	105	1013	700	19.7	9.3
2	0.14	0.29	0.48	109	105	111	113	833	573	17.7	7.6
4	0.28	0.57	1.10	133	133	101	99	680	407	12.9	5.4
7	0.50	1.00	1.88	153	146	136	133	547	313	11.1	4.4
10	0.70	1.41	2.68	153	151	138	141	533	280	11.3	4.4

* With acetic acid, 10% of $Zn(NO_3)_2 \cdot 6H_2O$.
† Contains 5.8% ethyleneurea; catalyst control fabric.

aldehyde values for untreated and catalyst control samples were essentially nil.

Results and Discussion

The properties of the three series of treated fabrics are listed in Tables I, II, and III.

In all three series, the catalyst control fabric has lower breaking and tearing strengths than the untreated fabric. This decrease is due to cellulose degradation caused by the acidic catalyst. The greatest strength loss occurs in the catalyst control for the formaldehyde series, while smaller (but still noticeable) losses occur in the control fabrics for the other two series. That the differences are due to different extents of cellulose degradation is evidenced by the data in Table IV. The degree of polymerization of cellulose in the fabric, determined by the fluidity of cupriethylenediamine solution [1], is reduced more by treatment with zinc nitrate alone, as in the catalyst control for the formaldehyde series, than by treatment with zinc nitrate in the presence of the nitrogenous compounds. Presumably the neutralizing effect on the acidic salt occurs throughout the series treated with dimethylol urea and dimethylol ethyleneurea although its extent may be modified by the influence of the methylol substituent on the basicity of the amide. The effect of acidic degradation and neutralization of the acidity by the amido groups had been noted previously with respect to the tearing strength of cotton fabric treated with dimethylol ethyleneurea [3], but a greater effect was reported than is observed in the present treatments.

TABLE II. Properties of Fabrics Treated with Dimethylol Urea

Treating solution		Fabric analysis		Fabric properties							
				Crease recovery angle, degrees		Wet crease rec. angle, degrees		Tearing strength, g.		Breaking strength, kg., W	Elongation at break, %, W
% DMU	% $Zn(NO_3)_2 \cdot 6H_2O$*	% N	% CH_2O	W	F	W	F	W	F		
0	0.00	—	—	91	84	93	94	1287	840	22.3	10.9
0†	0.70	—	—	93	79	86	106	1060	733	20.9	10.6
1	0.07	0.17	0.19	92	86	88	91	1127	687	21.9	9.5
2	0.14	0.30	0.33	100	96	86	83	1053	687	19.7	9.5
4	0.28	0.54	0.85	116	111	107	107	813	600	18.3	8.4
6	0.42	0.89	1.40	134	123	119	116	727	500	14.6	6.4
8	0.56	1.26	2.03	142	138	131	123	628	380	13.0	5.7
10	0.70	1.57	2.45	150	145	135	128	633	387	12.7	5.5
12	0.84	2.14	3.54	157	150	140	141	507‡	300	12.9**	4.6
15	1.05	2.65	4.33	158	152	138	136	513‡	293	13.1**	4.5

* With acetic acid, 10% of $Zn(NO_3)_2 \cdot 6H_2O$.
† Contains 5.0% urea; catalyst control fabric.
‡ For these fabrics, tearing strength (g.) of untreated fabric, 1107 W, 853 F; of catalyst control fabric, 993 W, 733 F.
** For these fabrics, breaking strength (kg.) of untreated fabric, 23.8; of catalyst control fabric, 22.3.

TABLE III. Properties of Fabrics Treated with Formaldehyde

Treating solution		Fabric analysis	Fabric properties							
			Crease recovery angle, degrees		Wet crease rec. angle, degrees		Tearing strength, g.		Breaking strength, kg., W	Elongation at break, %, W
% CH_2O	% $Zn(NO_3)_2 \cdot 6H_2O$	% CH_2O	W	F	W	F	W	F		
0	0.00	—	79	80	64	82	1053	807	22.4	11.2
0*	0.50	—	101	86	89	97	653	493	16.6	8.3
1	0.50	0.10	103	104	78	98	627	433	16.1	7.7
2	0.50	0.26	119	114	98	109	547	373	14.2	7.1
5	0.50	0.70	129	125	106	122	500	320	12.7	6.1
10	0.50	1.00	134	131	120	140	480	300	11.3	6.0
15	0.50	1.73	134	130	124	131	447	287	12.1	4.6
20	0.50	1.62	136	135	131	140	460	273	12.2	5.2

* Catalyst control fabric.

TABLE IV. Effect of Catalyst on Degree of Polymerization of Cellulose

	Treating solution	Degree of polymerization
DMEU blank fabric	No treatment	3870
DMEU catalyst control	0.7% $Zn(NO_3)_2 \cdot 6H_2O$ 0.07% acetic acid 5.8% ethyleneurea	3150
DMU blank fabric	No treatment	4050
DMU catalyst control	0.7% $Zn(NO_3)_2 \cdot 6H_2O$ 0.07% acetic acid 5.0% urea	4030
CH_2O blank fabric	No treatment	3880
CH_2O catalyst control	0.5% $Zn(NO_3)_2 \cdot 6H_2O$	1470

Calculation of the Degree of Cross-Linking

To correlate fabric properties with degree of cross-linking, the latter quantity was calculated from the nitrogen and formaldehyde analyses. It is expressed in this article as cross links per anhydroglucose unit (agu). This expression gives a value equal to half the fraction of anhydroglucose units connected to a cross-linkage on the assumption that no unit is connected to more than one cross-linkage.

With the series of fabrics treated with formaldehyde, the cross links per agu were equated with the moles formaldehyde per agu calculated from the percentage value obtained in the formaldehyde determination. This is based on the assumption that the cross links contain no more than a single methylene group and that there are no formaldehyde residues on the fabric that are not part of a cross link. These assumptions are supported by the work of Wagner and Pacsu [6] and Gruntfest and Gagliardi [4], although the latter workers report that longer formaldehyde cross links are formed at levels of substitution higher than encountered in this work.

Less simple calculations must be made with the series of fabrics treated with dimethylol ethyleneurea and dimethylol urea, as both of these compounds have been reported to form multiple unit cross links [3, 5]. For both treatments, it was assumed that the cross links had the structure shown in Figure 1, where the free valence bonds on the nitrogens are occupied by ethylene groups or hydrogen atoms. The substitution in cross links per anhydroglucose unit is then given by the following formulas.[2]

Cross links per agu = moles formaldehyde per agu $-\frac{1}{2}$ (moles nitrogen per agu)

$$\text{Moles formaldehyde per agu} = \frac{\%CH_2O \times 5.40}{100 - \left(\%N \times \frac{a}{28}\right) - \left(\%CH_2O \times \frac{14}{30}\right)}$$

$$\text{Moles nitrogen per agu} = \frac{\%N \times 11.57}{100 - \left(\%N \times \frac{a}{28}\right) - \left(\%CH_2O \times \frac{14}{30}\right)}$$

where $a = 84$ for the dimethylol ethyleneurea treatments and $a = 58$ for the dimethylol urea treatments. Calculations by these formulas eliminate from consideration any material containing two atoms of nitrogen for each formaldehyde residue. This composition is the probable one for material that remains on the fabric after washing but does not form cross links, such as bound or unbound polymer and singly bound urea or ethyleneurea groups. The presence of free methylol groups is discounted, as it is believed they are too labile to remain through the curing and washing procedures. In the case of the dimethylol urea treatments, the calculations also assume the absence of branched cross links through the remaining amido hydrogens and therefore may be less exact than those for the dimethylol ethyleneurea treatments. The results of the above calculations are shown in Table V. In addition to the values for cross link substitution, the ratio of moles formaldehyde residues to half the molar nitrogen content is given This value, which is the ratio of formaldehyde to urea units in Figure 1, is included to indicate the average size of cross link formed with the dimethylol ureas. It does not, however, neglect the bound but not cross-linking material as the value for cross link substitution does.

From the data in Table V it is noticed that cross-linking with the dimethylol ureas is inefficient at low add-ons. The low value of the $CH_2O:N/2$ ratio and low degrees of substitution indicate either a high proportion of bound material that does not form cross links or the formation of relatively long cross links. This is particularly evident with dimethylol urea,

[2] These formulas were derived from the general formula:

$$\text{Moles per agu} = \frac{\text{percent } CH_2O \text{ or N}}{100 - \text{percent urea residue} - \text{percent methylene}} \times \frac{\text{molar weight agu}}{\text{molar weight } CH_2O \text{ or N}}$$

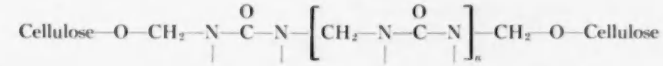

Fig. 1. Structure of cross link formed by dimethylol urea or dimethylol ethyleneurea.

where a relatively high nitrogen content is reached before appreciable cross-linkage occurs. Both dimethylol ethyleneurea and dimethylol urea tend to reach a constant-size cross link above the lowest add-ons. With dimethylol ethyleneurea, cross links with an average of 1.3–1.4 ethyleneurea residues are obtained. (In Figure 1, $n = 0.3$–0.4.) With dimethylol urea, cross links with an average of about 2.0 urea residues are obtained ($n = 1.0$). These sizes are approximately the same as previously reported for these two cross-linking agents [3, 5].

Relation of Wrinkle Resistance to Degree of Cross-Linking

From the previous data, the plot of warp crease recovery angle vs. cross link substitution for the three cross-linking agents, Figure 2, was prepared. In this plot, the values for zero substitution were taken from the catalyst control samples. It is observed that all three cross-linking agents produce maximum crease recovery angles at a substitution of 0.04–0.05 cross links per anhydroglucose unit and no further change in crease recovery at higher substitutions. Assuming that no anhydroglucose units are connected to more than a single cross link, 8–10% of the total anhydroglucose units are bound by cross links when maximum crease recovery or wrinkle resistance is obtained. If the cross-linking reaction occurs only in the accessible regions, about 20% of the total cotton fiber, then approximately half the anhydroglucose units in these regions are bound by cross links at this substitution. With such an extent of chemical modification, it is not surprising that the physical properties of the fiber are drastically modified.

The curves obtained from the dimethylol ethyleneurea and dimethylol urea treatments in Figure 2 are very similar in spite of the longer cross links formed by dimethylol urea. A higher add-on of dimethylol urea, however, is required to produce the same crease recovery angles because longer and therefore fewer cross links are formed. The formaldehyde treatment, which produces the shortest cross links, does show a significantly lower maximum in crease recovery angles obtained. Although it is tempting to attribute this difference to the shorter cross link or other characteristics of the treatment, crease recovery angles up to 150° in the warp direction have been obtained at this laboratory from similar formaldehyde treatments in frequent instances.

Results similar to the previous are obtained

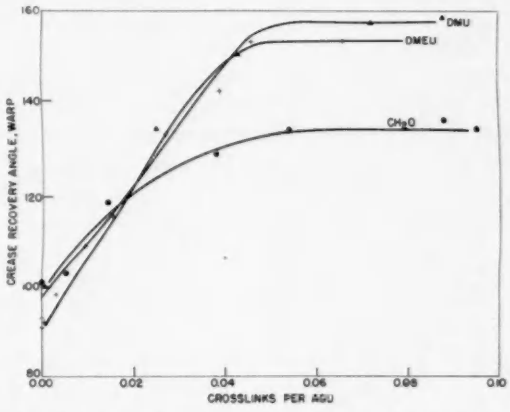

Fig. 2. Crease recovery angle of cross-linked fabrics.

TABLE V. Cross Link Substitution on Treated Cotton Fabric

Cross-linking agent	N, %	CH_2O, %	Cross links per agu	Molar ratio, CH_2O residues per $\frac{N}{2}$
Dimethylol urea	0.17	0.19	0.0004	1.05
	0.30	0.33	0.0005	1.05
	0.54	0.85	0.015	1.45
	0.89	1.40	0.025	1.46
	1.26	2.03	0.039	1.50
	1.57	2.45	0.043	1.46
	2.14	3.54	0.072	1.54
	2.65	4.33	0.087	1.53
Dimethylol ethyleneurea	0.11	0.16	0.003	1.35
	0.29	0.48	0.009	1.55
	0.57	1.10	0.027	1.80
	1.00	1.88	0.046	1.75
	1.41	2.68	0.066	1.77
Formaldehyde	—	0.10	0.005	—
	—	0.26	0.014	—
	—	0.70	0.038	—
	—	1.00	0.054	—
	—	1.62	0.088	—
	—	1.73	0.095	—

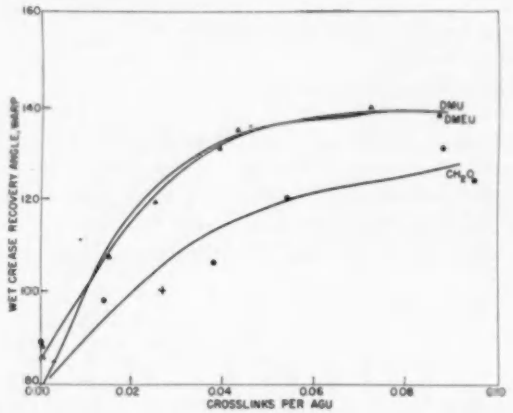

Fig. 3. Wet crease recovery angle of cross-linked fabrics.

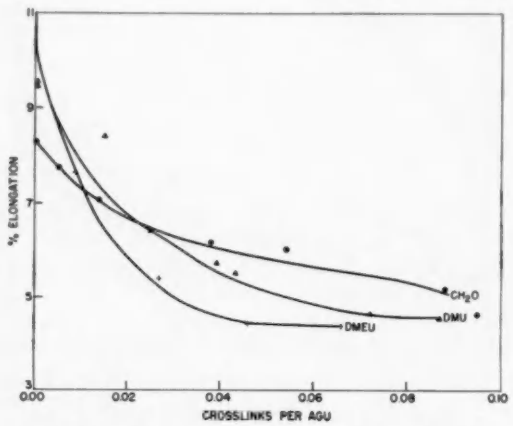

Fig. 4. Elongation at break of cross-linked fabrics.

by plotting wet crease recovery angles vs. cross link substitution as in Figure 3. The reproducibility of this test is poorer than that of the test with conditioned samples, but the results are sufficient to show that the curves obtained are similar to those of Figure 2. Maximum recovery angles are reached at about the same substitution but are lower than the dry or conditioned recovery angles.

Extensibility of Cross-Linked Fabric

One of the properties associated with wrinkle resistance in treated cotton fabric is a reduction in extensibility. The relation of this property to degree of cross-linking is shown by the plot of elongation at break vs. cross links per anhydroglucose unit shown in Figure 4. Again the zero substitution values are from the catalyst control samples.

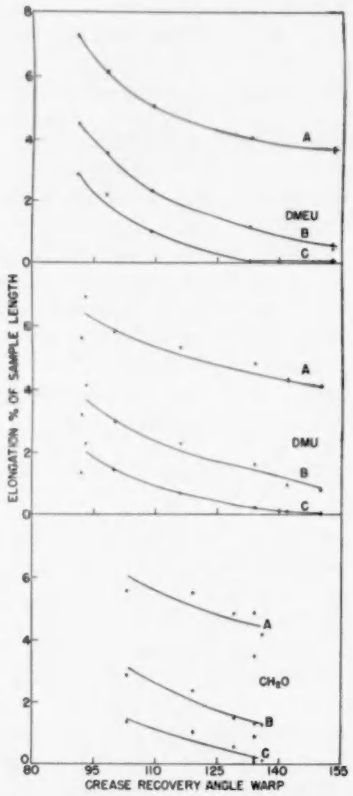

Fig. 5. Elongation (A), residual elongation after immediate recovery (B) and after 60 min. rest (C) of wrinkle-resistant fabrics submitted to a warp load of 8 kg.

Elongation at break, in Figure 4, is reduced rapidly as substitution increases initially, and more slowly or not at all as substitution increases in the higher ranges. With the dimethylol ethyleneurea and dimethylol urea treated fabrics, the latter stage is reached near the substitution at which maximum crease recovery angles are reached, 0.04–0.05 cross links per anhydroglucose unit. With the formaldehyde-treated fabric this stage is reached at a less definite substitution; as with crease recovery angles in Figure 2, a less sharp change with substitution occurs as an ultimate value is approached.

Relation of Elastic Properties to Wrinkle Resistance

It is common practice to use crease recovery angles as a measure of wrinkle resistance, as is done in this investigation. These recovery angles are actually a measure of the ability of the fabric to recover from deformation. This recovery from

deformation, rather than a resistance to deformation, has been shown to be the physical mechanism of wrinkle resistance [2]. The following part of this investigation, therefore, was conducted to show the relation of recovery from elongation to crease recovery angle.

For these tests, strips of the cotton fabrics containing 80 warp threads were used in the Instron Tester. Bench marks were made on the strips while in the jaws of the tester to mark the original 5-in. gauge length. The strips were subjected to an 8-kg. warp load, and the tester then was reversed, allowing the fabric strips to recover. The 8-kg. loading was chosen because it was sufficient to cause appreciable elongation and yet was well below the breaking load of all samples tested.

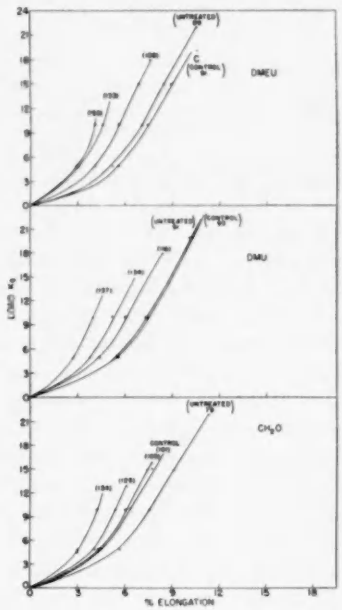

Fig. 6. Load–elongation curves of wrinkle-resistant fabrics. Value for warp crease recovery angles in parentheses.

From the curves obtained, the elongation at 8-kg. load and the elongation remaining after recovery to zero load were noted. The latter value is described as residual elongation after immediate recovery, and the difference between the two values is described as the immediate recovery. Sixty minutes after this test, the samples were again placed in the tester with the jaws at the bench marks and elongated. The elongation between the 5-in. gauge length and the zero position on the extrapolated elongation curve is described as permanent set. These values are all expressed as percent of original sample length.

In Figure 5 are shown plots of the elongation values vs. warp crease recovery angles for each of the three series. As in previous plots, the curves initiate with values from catalyst controls. Except for the smaller range of the plot from the formaldehyde series, the curves are very similar. The following characteristics are common to all three cross-linking treatments.

1. Elongation under 8-kg. load is reduced as crease recovery angle increases.

2. Immediate recovery increases slowly as crease recovery angle increases. Elongation remaining after immediate recovery decreases more rapidly because of the decrease in total elongation.

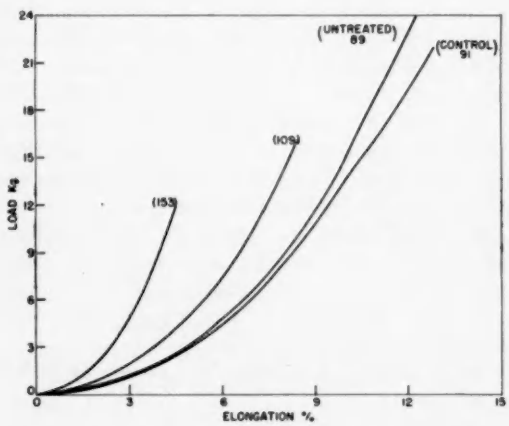

Fig. 7. Load–elongation curves of wet fabrics, DMEU treatment.

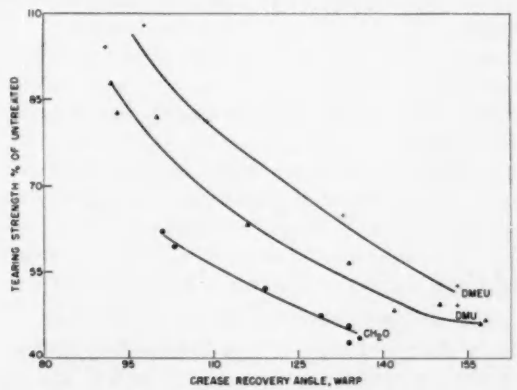

Fig. 8. Tearing strength of wrinkle-resistant fabrics, as percent tearing strength of untreated fabrics.

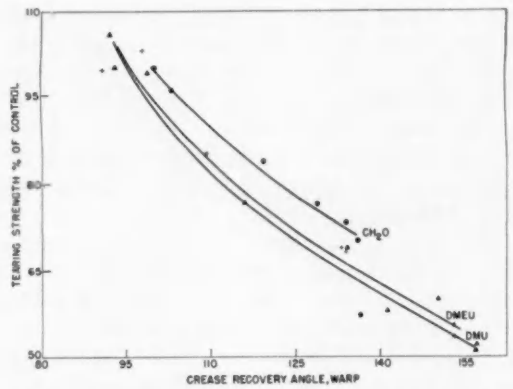

Fig. 9. Tearing strength of wrinkle-resistant fabrics, as percent tearing strength of catalyst control fabrics.

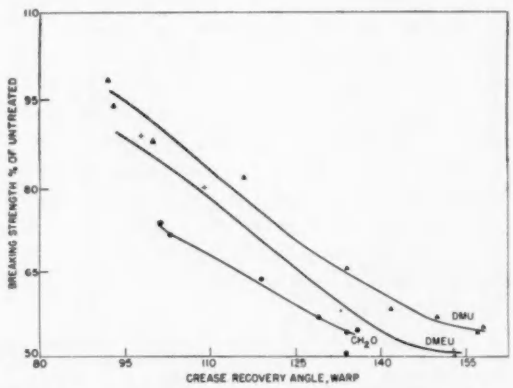

Fig. 10. Breaking strength of wrinkle-resistant fabrics, as percent breaking strength of untreated fabrics.

3. Permanent set decreases as crease recovery increases and approaches zero at 135° warp recovery angle.

4. Recovery during the 60 min. after immediate recovery is completed remains close to a constant value until permanent set approaches zero and then decreases.

5. Little or no change occurs in these elongation values after maximum crease recovery angle is attained. Therefore, after a substitution of 0.05 cross links per anhydroglucose unit is reached, further substitution gives no increase in elastic recovery.

The increase in elastic modulus of the fabric shown by the decreased elongation at 8 kg. is also exhibited by the increasing steepness of the load–elongation curves of the fabrics as cross link substitution increases. In Figure 6, load–elongation curves derived

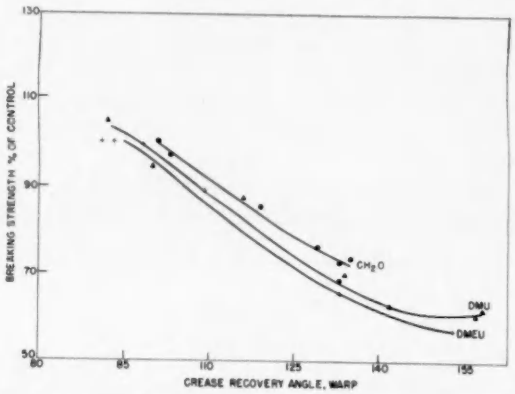

Fig. 11. Breaking strength of wrinkle-resistant fabrics, as percent breaking strength of catalyst control fabrics.

from Instron tracings are shown for selected samples from each series. Curves for the untreated and catalyst control fabrics are included for each of the series. In the dimethylol ethyleneurea and dimethylol urea series, these two curves differ little from each other. A greater, although still small, difference between these two curves is shown in the formaldehyde series. This difference and the somewhat higher crease recovery angles of the catalyst control for the formaldehyde series is apparently due to an effect similar to cross-linking caused by the action of zinc nitrate alone.

Load–elongation curves were also obtained from some fabric samples tested in the wet state instead of the conditioned. Selected samples from the series of dimethylol ethyleneurea-treated fabrics were wet and blotted, then tested in this condition to produce the curves shown in Figure 7. In this state, the untreated and catalyst control fabrics produce less steep curves than they do in the conditioned state, indicating a reduced modulus in the wet state. The cross-linked fabric with a high warp crease recovery angle, 153°, produces a curve identical with that produced in the conditioned state; this shows the stabilization produced by cross-linking. A fabric with a lower crease recovery angle, 109°, produces an intermediate curve with less change from that produced in the conditioned state than occurs in the curves produced by the untreated fabric.

Effect of Wrinkle Resistance Treatments on Fabric Strength

In the treatment of cotton fabric to produce wrinkle resistance with agents such as used in this in-

vestigation, there are two factors contributing to loss of strength. As noted previously, one is the cellulose degradation caused by the acidic catalyst. The other factor is the restriction of localized movements around the cross-linked sites, shown by the reduced extensibility or elongation at break of the cross-linked fabric. This restriction reduces the amount of possible stress distribution within the fibers and yarns of the fabric. The data in Tables I, II, and III indicate that the second factor is the major one. In the following part of this investigation the relation of tearing and breaking strength to crease recovery angles and the cross-linking agent was examined.

Figure 8 is a plot of crease recovery angles vs. Elmendorf tearing strength, both measured in the warp direction, for fabrics treated with the three cross-linking agents studied. In this figure, tearing strength is expressed as percent of the tearing strength of the untreated fabrics. These curves include values from the catalyst control fabrics but not those from the untreated fabrics. The graph shows that the formaldehyde treatment results in lower retention of tearing strength than the other two treatments at equal crease recovery angles, and the dimethylol ethyleneurea treatment results in greater retention of tearing strength than the dimethylol urea or formaldehyde treatments.

When tearing strengths are expressed as percent of the tearing strength of the catalyst control and plotted against crease recovery angles, the graph in Figure 9 is obtained. By this method, the curves are corrected for the different amounts of degradation caused by the catalyst, and the entire strength loss is due to the restriction of stress distribution. Now the dimethylol ethyleneurea and dimethylol urea treatments give essentially identical curves and the formaldehyde treatment gives a slightly higher tearing strength retention at equal crease recovery angles. The increased tearing strength of the formaldehyde-treated fabrics, if significant, may be due to a "repairing" effect of cross-linking on the degraded cotton fabric, or to a protective action by the initial cross-linking which reduces the total amount of acid degradation.

From the data presented graphically in Figures 8 and 9, it is concluded that the structure or size of the three cross links formed by the agents used has no influence on the tearing strength of the treated fabrics. Variations between the cross-linking agents in tearing strength retained at equal crease recovery angles are due to differences in the amount of cellulose degradation caused by the catalyst during treatment.

A similar examination was made of the breaking strength data obtained from the three series of cross-linked fabrics. In Figures 10 and 11 are the graphs obtained by plotting warp crease recovery angles vs. warp breaking strengths expressed as percent of the breaking strengths of untreated fabrics and catalyst control fabrics respectively.

A comparison of breaking strength as percent of untreated fabric at equal crease recovery angles shows that the formaldehyde treatment produces the lowest strength, the dimethylol ethyleneurea the intermediate strength, and the dimethylol urea the highest strength. The comparison of the three series with respect to breaking strength as percent of breaking strength of catalyst control fabrics, Figure 11, gives results similar to those obtained with tearing strengths. Fabrics treated with the three cross-linking agents have nearly the same strengths at equal crease recovery angles, although the formaldehyde treatment gives slightly greater strengths than do the other treatments.

As with tearing strength, all differences in breaking strengths at equal crease recovery angles that exist between treatments with these three cross-linking agents are explainable by differences in cellulose degradation caused by the acidic catalyst; none can be attributed to differences in the structure or size of the cross link formed.

Formaldehyde has an economic attractiveness for commercial use as a wrinkle-resistance finishing agent for cotton fabrics and probably would be so used if the strength losses, as shown in Figures 8 and 10, could be reduced. Such strength losses, however, appear to be intrinsic to the formaldehyde treatment. The higher strength losses obtained with formaldehyde, as compared with the methylol ureas, in this investigation are due to the high acidity of the catalyst system used; such acidity is apparently necessary to promote the cross-linking reaction of formaldehyde.

Summary and Conclusions

An examination of cotton fabrics treated with three cross-linking agents, dimethylol ethyleneurea, dimethylol urea, and formaldehyde, to obtain wrinkle resistance has led to the following conclusions.

With any of the three cross-linking agents, maximum crease recovery angles are obtained at a substitution of 0.04–0.05 cross links per anhydroglucose unit. Treatments with dimethylol ethyleneurea and dimethylol urea have a nearly identical relation of crease recovery angle to cross link substitution. Treatment with formaldehyde, however, gave a maximum recovery angle smaller than the maximum obtained with the other two agents.

Values for other physical properties of the treated fabrics also tend to approach a maximum deviation from those of the untreated fabric at the substitution where maximum crease recovery angles are obtained, 0.04–0.05 cross links per anhydroglucose unit.

Elongation of the fabric under a given load, 8 kg. for a strip containing 80 warp threads, is decreased as crease recovery angle increases. Permanent set with this loading decreases as crease recovery angles increase and approaches zero when a warp crease recovery angle of 135° is obtained, at a substitution of approximately 0.03 cross links per anhydroglucose unit. Immediate recovery, as percent of total length, increases as substitution increases. The amount of delayed recovery in the next 60 min. remains nearly constant until permanent set approaches zero, whereafter the delayed recovery decreases with increasing substitution until maximum crease recovery angles are reached.

Load–elongation curves of the treated fabrics show increasing steepness, indicative of increasing modulus, as cross link substitution increases. Cross-linked fabrics tested in the wet state produce load–elongation curves that are essentially the same as those produced by the dry fabric, although untreated fabric produces a curve of reduced steepness in the wet state.

Two factors contribute to strength loss in wrinkle-resistance treatments of cotton fabric: reduction of extensibility and subsequent restriction of stress distribution due to cross-linking, and acid degradation of the cellulose by the acidic catalysts. The first of these is the major factor, but the second also causes increased loss of tearing and breaking strengths particularly in the formaldehyde treatments.

Differences in the amount of tearing or breaking strength, retained by fabrics treated to give the same degree of wrinkle resistance by these cross-linking agents, are due to different amounts of cellulose degradation occurring in the application of the agents. None of the differences in strength retained is due to the size or structure of the cross links formed. The much higher strength losses in the formaldehyde treatment are caused by the higher acidity of the catalyst system used.

Acknowledgments

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Properties of Cotton Containing Radiation-Polymerized Acrylonitrile[1]

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COTTON, a versatile fiber, maintains its primary position in textile per capita consumption in the United States. In recent years, chemical modifications of cotton fiber to give it new and desirable properties have become major factors in cotton utilization [11]. During the investigation of the mechanism of radiation-induced reactions in cotton cellulose, it was noted that massive dosages of high energy gamma radiation induced major structural changes in the cotton cellulose molecule [1, 7–9]. However, at lower dosages, the changes were small, and the tensile strengths of the cotton fibers were not significantly lowered. The use of lower dosages of radiation to polymerize unsaturated monomers in the presence of cotton was indicated. Further, if the monomer could be polymerized inside the cotton fiber, this should give a fiber with new properties.

Initially acrylonitrile, a monomer which is easily polymerized by radiation [2, 3], was used. In this report, results of investigations to radiation-polymerize acrylonitrile in the presence of cotton and properties of the modified cotton are discussed.

Experimental

Radiation Source

The gamma irradiations were performed at the Southern Regional Research Laboratory by a distributed, kilocurie cobalt^{60} source. The dose rate of the source was about 0.6×10^6 roentgens per hour (maximum).

Materials

Deltapine cotton was spun into a 7s/3 yarn with a balanced mercerizing twist [10] and then wound into 18-yd. skeins weighing 4 g. The cotton was purified by extraction with hot ethanol and by boiling for 2.5 hr. in 1% NaOH solution, kept fresh by continuous removal and addition of NaOH solution with proper precautions to prevent exposure to air [12]. The NaOH was removed from the cotton by washing with distilled water, souring with dilute acetic acid, neutralizing with dilute NH_4OH, and then a second washing with distilled water. The cellulose was then allowed to condition at 21° C. and 65% relative humidity.

A commercial grade of acrylonitrile (water saturated), reagent grade $ZnCl_2$, and reagent grade N,N-dimethylformamide solvent were used.

Methods

Solutions of acrylonitrile in aqueous $ZnCl_2$ were uniformly padded on the yarns to give about a 200% pick-up, using about 100 g. of solution to 4 g. of yarn. For example, a typical treating solution contained 32 parts of acrylonitrile in 68 parts of aqueous 80% $ZnCl_2$. The treated yarns were then irradiated to give the desired dosage. Monomer was removed by washing the irradiated yarns with distilled water; loosely bound polymer was removed by extraction of the yarns overnight at room temperature with N,N-dimethylformamide in the ratio of about 4 g. of yarn to 75 ml. of polymer solvent, followed by washing with water and drying overnight at room temperature.

The content of nitrogen was determined using the Kjeldahl method [4]. The content of polymer was calculated from the unextractable nitrogen content. The moisture regain data were obtained by equilibrating the samples under standard conditions (21° C. and 65% relative humidity) from the dry side. The samples were then dried for 5 hr. in a forced draft oven at 105° C. The percent moisture, based on the oven-dry weight of the sample, was reported as moisture regain [6]. Yarn densities were determined with a gradient column as described

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TABLE I. Effect of Reagent Concentration on Radiation Polymerization of Acrylonitrile onto Cotton and on its Properties*

Property of yarn†	Acrylonitrile, 32 parts; Aqueous $ZnCl_2$, 68 parts of 65%	75%	80%	80%‡	Aqueous $ZnCl_2$, (75%), 80 and 60 parts, resp.; Acrylonitrile, parts 20	40
Polymer content, %	0.3	4.9	22.8	0.0	3.7	6.3
Breaking strength, lb.	10.1	8.6	8.7	9.8	8.9	8.2
Elongation-at-break, %	13.5	15.6	23.2	13.9	18.6	14.9
Yarn number, tex	252	281	379	248	286	283
Breaking stress, g./tex	18.1	13.9	10.4	17.9	14.1	13.2
Breaking toughness, g./tex	0.77	0.69	0.80	0.74	0.86	0.60
Average stiffness, g./tex	134	89	45	129	76	88

* Radiation dosage, 0.8 megaroentgen.
† Instron properties.
‡ Treated, unirradiated yarn.

TABLE II. Effect of Radiation Dosage on Polymerization of Acrylonitrile onto Cotton and on its Properties*

Property of yarn†	Dosage, megaroentgen 0.0‡	0.0**	0.2	0.4	0.6	0.8	1.0
Polymer content, %	0.0	0.0	3.6	12.3	15.9	26.0	25.8
Breaking strength, lb.	10.1	9.8	8.7	8.5	9.1	8.2	8.8
Elongation-at-break, %	13.5	13.9	12.6	13.9	16.5	24.6	22.9
Yarn number, tex	241	248	256	276	300	398	401
Breaking stress, g./tex	19.0	17.9	15.3	14.0	13.7	9.3	9.9
Breaking toughness, g./tex	0.79	0.74	0.58	0.63	0.73	0.70	0.73
Average stiffness, g./tex	141	129	121	101	83	38	43
Moisture regain, %	7.6	7.7	7.5	7.1	7.1	6.5	6.4

* Treating solution: 32 parts of acrylonitrile in 68 parts of aqueous 80% $ZnCl_2$.
† Instron properties.
‡ Untreated, unirradiated yarn.
** Treated, unirradiated yarn.

by Orr et al. [13]. After ten-fold dilution of the treating solutions, pH values were determined, 1 and 5 *M* acetic acid being added in the place of water in the solvent to adjust the pH of the solution from an initial value of 5.2 to 3.8 and 2.6 respectively. Textile tests were done according to ASTM methods [5]. The IP-4 inclined plane tester[3] was operated at a constant time to break of approximately 20 sec. A gauge length of 10 in. and a specimen rate of extension of 50%/min. were used with the Instron.[3] Optical and electron microscopical observations were made using previously described techniques [14].

Results

Reagent Concentration

The effects of reagent concentration on the radiation polymerization of acrylonitrile onto cotton and on its properties are shown in Table I. By use of a reagent concentration of 32 parts of acrylonitrile in 68 parts of aqueous 80% $ZnCl_2$, and at a massive radiation dosage of 0.8 megaroentgen, the maximum add-on of unextractable polymer was obtained. Under these conditions, the breaking strength of the yarn decreased about 15%; the breaking stress, about 45%; and the average stiffness, about 65%. The elongation-at-break increased about 70%; and the yarn number, about 55%. The breaking toughness remained about constant.

Radiation Dosage

The effects of radiation dosage on the polymerization of acrylonitrile onto cotton and on its properties, using the optimum treating solution, are shown

[3] Trade names are given as part of the exact experimental conditions and not as an endorsement of the products over those of other manufacturers.

in Table II. The conversion of monomer to unextractable polymer is related to the total dosage, as shown previously [2, 3] and as indicated by polymer add-on. It is observed that the total dosage may vary between 0.8 and 1.0 megaroentgen, with the treated yarns produced at each dosage having about the same properties. It is noted that the moisture regain decreased with increasing dosage and polymer add-on.

TABLE III. Effect of pH of Treating Solution on Radiation Polymerization of Acrylonitrile onto Cotton and on its Properties*

Property of yarn‡	pH of treating solution† 2.6	3.8	5.2
Polymer content, %	25.0	25.7	24.6
Breaking strength, lb.	6.5	8.1	9.3
Elongation-at-break, %	12.8	17.8	19.2

* Radiation dosage, 1.0 megaroentgen.
† Treating solution: 32 parts of acrylonitrile in 68 parts of aqueous 80% $ZnCl_2$; pH adjusted by substitution of water by acetic acid in $ZnCl_2$ solution; pH measurement made on solvent after ten-fold dilution.
‡ IP-4 properties.

TABLE IV. Effect of Heating on Properties of Treated Yarn*

Yarn treatment	Time of heating at 165° C., min.	Breaking strength, lb.	Elongation-at-break, %
Untreated control	0	11.3	11.0
Polymer content 28.3%	0	8.7	19.5
Polymer content 27.2%	15	8.3	17.1
Polymer content 26.8%	30	7.9	15.4
Untreated control	30	9.7	10.2

* Treating solution: 32 parts of acrylonitrile in 68 parts of aqueous 80% $ZnCl_2$; 0.8 megaroentgen. IP-4 properties.

pH of Treating Solution

As the acidity of the treating solution increased, the breaking strength and elongation-at-break of the treated yarns decreased for the same radiation dosage and polymer add-on, as shown in Table III.

Heating of Treated Yarns

The resistance of treated (acrylonitrile solution added and then yarn irradiated) yarns to the effects of heating at 165° C. is shown in Table IV. In the untreated (purified) yarn, decreases in breaking strength and in elongation-at-break with increased time of heating are noted. The treated yarns, containing approximately the same polymer add-on, showed similar effects on heating; however, after being heated the treated yarn still showed an increase of about 50% in elongation-at-break as compared with the untreated yarn.

Acid Resistance

Treated yarns, containing about 25% polymer, were about as acid resistant as untreated yarns. For example, after untreated yarn was immersed in 50% aqueous sulfuric acid at room temperature for 30 min., its breaking strength decreased about 40% and its elongation-at-break, about 15%. Similarly with treated yarn, the decrease in breaking strength was about 35% and in elongation-at-break, about 25%.

Comparison of Yarn Treatment

In Table V, a comparison of the effects of treatment of yarn on its properties is made. It is readily

TABLE V. Comparison of the Effects of Treatment of Yarn on its Properties*

Property of yarn†	Yarn treatment: Raw	Purified	Purified irradiated	Treated	Treated irradiated
Polymer content, %	—	—	—	—	24.0
Breaking strength, lb.	10.0	9.2	8.7	9.6	8.6
Elongation-at-break, %	13.7	12.5	12.3	13.1	23.4
Yarn number, tex	260	240	246	241	386
Breaking stress, g./tex	17.5	17.4	16.0	18.1	10.2
Breaking toughness, g./tex	0.82	0.71	0.65	0.75	0.80
Average stiffness, g./tex	128	139	130	138	44

* Where indicated, treating solution: 32 parts of acrylonitrile in 68 parts of aqueous 80% $ZnCl_2$, radiation dosage: 0.8 megaroentgen.
† Instron properties.

TABLE VI. Properties of Treated Yarn*

Yarn treatment	Breaking strength,† lb.	Polymer content, %	Yarn density, g./ml.
Untreated control	9.9	0.11	1.5333
0.4 megaroentgen	9.1	13.8	1.462
0.6 megaroentgen	8.8	19.2	1.434
0.8 megaroentegn	9.7	24.6	1.421

* Treating solution: 32 parts of acrylonitrile in 68 parts of aqueous 80% $ZnCl_2$.
† IP-4 properties.

seen that the polymer add-on and the radiation dosage are the major factors affecting the properties of the yarn. Comparison of data in Table V with data in Tables I and II indicates that the effects noted are fairly reproducible. Fibers were easily separated from the differently treated yarns. It was determined that the treatment affected the properties of the fibers.

Yarn Density

The densities of the yarns are proportional to the polymer add-on, the density of the polymer being less than that of the yarn, as shown in Table VI. The breaking strength would indicate only a small change in this property of the yarns due to radiation dosage, ranging from 0.0 to 0.8 megaroentgens.

Location of the Polymer

Ultrathin sections of purified cotton fiber containing radiation-polymerized acrylonitrile, about 25% polymer content, were examined by electron microscopy to determine the location of the polymer in the fiber. The electron micrographs, shown in Figure 1, indicate that the polymer is located primarily within the fiber in an area well within the secondary wall about halfway between the fiber edge and the lumen.

Figure 1A shows the polymer located between the outer edge of the fiber and the lumen. The cellulose fiber wall was then removed by extraction with 0.50 *M* cupriethylenediamine for 30 min.; then the extracted sample was placed in N,N-dimethylformamide overnight to dissolve the polymer. Figure 1B shows the insoluble polymer residue remaining after removal of the cellulose wall and any readily soluble polymer. Figure 1C shows the small amount of the cellulose wall remaining around the polymer after extraction with 0.50 *M* cupriethylenediamine for 30 min. Figure 1D is the same as Figure 1C except at higher magnification. Figure 1D indicates that some polymer is also located within the outer wall of the fiber.

Summary and Conclusions

The modification of the physical properties of cotton fiber, by exposing the fibers in the presence of acrylonitrile monomer, has been accomplished. Relatively large quantities of acrylonitrile monomer in aqueous zinc chloride were applied to cotton and radiation-polymerized in the presence of the cotton. Microscopic examination indicated that the polymer was located within the fiber. There was only a slight decrease in breaking strength, a significant increase in elongation-at-break, and a decrease in stiffness of the fibers. Resistance to acid and to heat degradation of the yarn containing polyacrylonitrile was comparable to that of untreated yarn. A radiation dosage of 0.8 megaroentgen gave a maximum add-on of polymer.

The results are particularly interesting relative to the preparation of chemically modified cottons. Work will be continued to investigate the application of additional chemicals to cotton by high energy radiation and to evaluate the properties of these chemically modified cottons.

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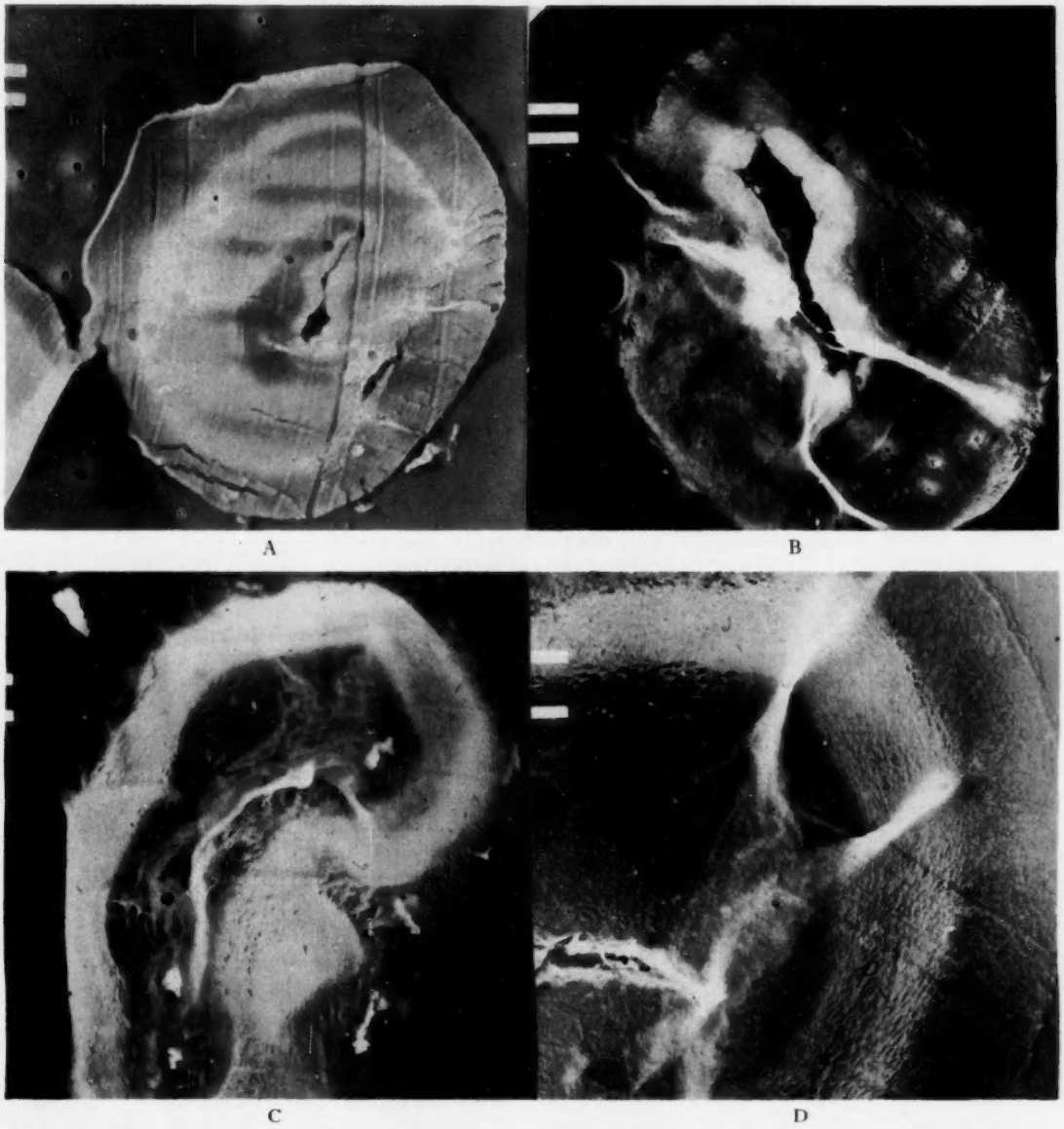

Fig. 1. Electron micrographs of ultrathin sections of purified cotton fiber containing radiation-polymerized (0.8 megaroentgen) acrylonitrile (26% polymer content). (A) Location of polymer between lumen and outer cellulose wall; 5000 ×. (B) Polymer remaining after outer wall is removed by extraction with cupriethylenediamine; then fiber is placed overnight in N,N-dimethylformamide; 5000 ×. (C) Polymer remaining after outer wall is removed by extraction with cupriethylenediamine; 5000 ×. (D) same as (C); 10,000 ×.

Effects of Gamma, High-Energy Electron, and Thermal Neutron Radiations on the Fibrillar Structure of Cotton Fibers

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Abstract

Changes in the structure of cotton fibers after exposure to gamma, high-energy electron, and thermal neutron radiations, in various ambient atmospheres, were evaluated by the techniques of optical and electron microscopy. Radiation exposures evaluated were: gamma rays, 10^5 to 4×10^8 r.; 2-Mev electrons, 5×10^4 to 10^8 rep; integrated thermal neutron fluxes, 5×10^{14} to 10^{17} nvt. Changes in the physical and chemical properties of similarly irradiated fibers have been, or are to be, reported in other publications. All irradiated, unmodified cottons could be differentiated from unexposed cottons by optical microscopical observations of samples stained in Nile Blue Sulfate. Gamma ray and electron exposed samples differed from unexposed fibers in swelling behavior in 0.2 *M* (in Cu) cupriethylenediamine hydroxide, but thermal-neutron-irradiated cotton differed from the control only after exposure to an integrated flux of 10^{17} nvt. Electron micrographs of the fibrillation patterns exhibited by fibers exposed to gamma and electron radiation doses of 5×10^6 to 10^8 r. or rep on beating in water in a Waring Blendor showed the formation of increasingly smaller fragments of samples. Some shortening of fibrils and an increase in number of broken ends of microfibrils were seen in neutron-irradiated cotton exposed to 10^{17} nvt; at lower integrated neutron fluxes, fibrillation was unchanged. No distinct differences were found in microscopical tests between gamma and electron irradiated fibers or between gamma-exposed fibers irradiated in oxygen and nitrogen. At doses of 4×10^6 r. of gamma rays, the trend toward smaller fragments reversed and short chunks of material were observed.

Varying amounts of degradation were observed in modified cottons after irradiation. Smaller fragments were observed in cottons modified to change and reduce the total crystalline cellulose; the fibrillation of acrylonitrile-treated cotton appeared unchanged, but irradiation produced a decrease in the size of fragments of dichloropropanol- and dimethylol cyclic ethyleneurea-modified samples.

Introduction

Early investigations of cottons exposed to high-energy radiations showed that scission of the chains of the fiber cellulose was the predominant result of reaction with radiation [19]. The decrease in degree of polymerization of cotton cellulose was found equal to that of wood cellulose, and it was concluded that the fine structure of cotton cellulose is homogeneously accessible to the radiations [6]. More recent studies by Blouin and Arthur [3, 4] of purified lint cottons irradiated with gamma rays showed no changes in the crystallinity of irradiated samples as estimated by infrared absorption and x-ray diffraction techniques, but marked increases in the rates of acid hydrolysis were observed. These authors concluded that irradiation results in the oxidation of the cotton cellulose. The physical and chemical properties of irradiated cottons have been investigated also by Pan and his associates [16, 17], who studied cottons exposed to high-energy electrons; by Teszler [22, 23, 24], who investigated the effects of gamma and neutron irradiation of cotton cellulose; and by Gilfillan and Linden [12, 13], who employed three types of high-energy radiations.

The studies reported in this paper were made to examine the possibilities of analyzing microscopically the changes in fibrillar organization of the cotton fiber after high-energy irradiation. The optical

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microscope was employed to study, by means of the usual staining and swelling techniques, the effect of each type of irradiation on whole fibers. Electron microscopy enabled observations of changes in microfibrillar structure. Untreated cottons and cottons purified by the extraction techniques described in ASTM, D539-57T [2] were observed after irradiation by gamma rays, 2-Mev electrons, and thermal neutrons. Additional observations were made on partially acetylated cotton fabrics and fabrics which had been treated with dichloropropanol, dimethylol cyclic ethyleneurea, or acrylonitrile following doses of 5×10^6 and 10^7 r. of gamma radiation.

Irradiation Procedures

Gamma

Samples of the gamma-irradiated, purified cottons studied by Blouin and Arthur [3] were compared with small samples of unpurified sliver, simultaneously irradiated for microscopical studies only. These cottons were irradiated in oxygen and nitrogen atmospheres by gamma radiations from fission products of the spent fuel elements of the Materials Testing Reactor (MTR) at the National Reactor Testing Station, Idaho Falls, Idaho. Other gamma-irradiated samples were obtained by exposure in the Co^{60} facility at the Southern Regional Research Laboratory. Cumulative doses of gamma rays ranged from 10^5 to 4×10^8 r. (roentgens).

Electron

Cottons exposed to high-energy electron beams were obtained from the samples studied by Pan and his associates [16]. These unpurified cottons were moisture conditioned at 70° F. and 65% RH, then irradiated in ambient laboratory conditions with 2-Mev electrons from a Van de Graaff accelerator. Cumulative doses of monoenergetic (2 Mev) electrons ranged from 5×10^4 to 10^8 rep (roentgens equivalent physical).

Neutron

Neutron-irradiated samples consisted of purified cottons exposed in the thermal neutron column of the Materials Testing Reactor at Idaho Falls, Idaho, for studies by Blouin and Arthur [5].

Integrated fluxes of thermal neutrons ranged from 5×10^{14} to 10^{17} nvt (thermal neutrons/cm.^{2}/sec. $\times$ exposure time in seconds).

Reaction of Irradiated Cotton with Dyes and Alkaline Swelling Agents

Absorption of a Basic Dye and Swelling in Cuprietylenediamine Hydroxide

Nile Blue Sulfate (Nile Blue A, C.I. 51180) is a basic dye of the oxazine series which, when used in a neutral solution, attaches to acidic groups. In unpurified, native cotton fibers the noncellulosic materials in the primary wall and lumen are stained dark blue [15]. Unmodified, purified cottons stain a lighter blue, but modifications resulting in carboxyl group formation cause increased depth of staining.

In all irradiated samples, staining tests with Nile Blue Sulfate showed some increased absorption of the dye, the depth of color increasing with the cumulative exposures. In the gamma-irradiated samples, no distinct differences were observed between fibers irradiated in atmospheres of oxygen or nitrogen. Samples of unpurified yarns exposed to electron beams showed somewhat less absorption of the dye at numerically equivalent dose levels; the neutron-irradiated samples, while they could be differentiated

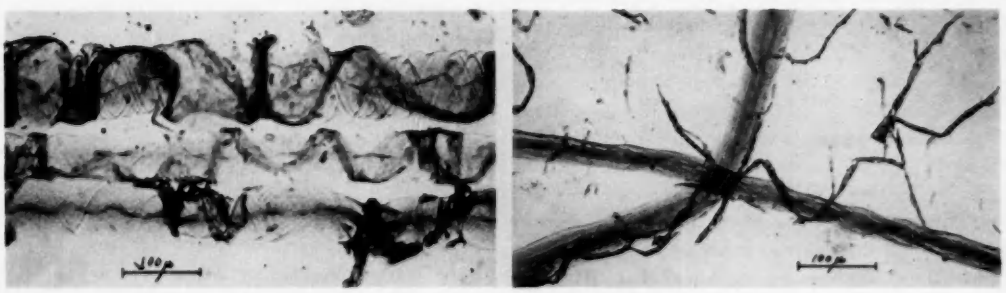
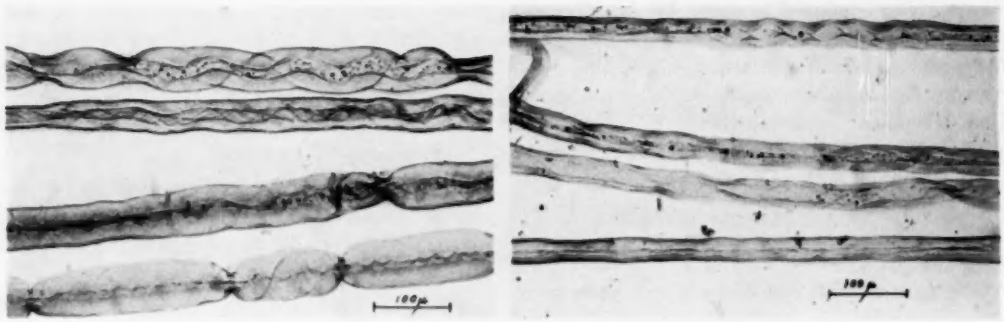


Fig. 1. Photomicrographs of swelling of unpurified irradiated cotton in cuene. (A) Control; (B) gamma-irradiated, 10^6 r., O_2 atmosphere.



A B

Fig. 2. Photomicrographs of swelling of purified irradiated cotton in cuene. (A) Control; (B) neutron-irradiated, 10^{17} nvt, in air.

from unexposed samples, apparently absorbed much smaller amounts of dye than gamma- or electron-irradiated samples.

In the practice of fiber microscopy, cupriethylenediamine hydroxide (cuene), a cellulose solvent, is used in low concentrations to swell cellulose fibers to permit observation of the internal structure in the optical microscope. Solutions approximately 0.2 *M* in Cu swell undegraded fibers extensively and produce balloon- or sausage-shaped structures (Figure 1A). Degraded fibers, or those in which the cellulose has been modified to produce alkali-sensitive links, dissolve more quickly in reagents of this concentration without formation of balloon or sausage structures.

The swelling behavior of all gamma-irradiated fibers readily differentiated irradiated from unexposed fibers. In addition to the absence of balloon or sausage formations in the exposed fibers, some purified fibers dissolved during observation after a dosage of only 10^5 r., and increasing numbers dissolved in samples receiving larger doses. Although some unpurified fibers dissolved after doses of 10^7 r. and 5×10^7 r., no dissolution was observed in samples receiving 10^8 r. At this latter dose, the formation of carboxyl groups in the noncellulosic materials and in the cellulose may have prevented the formation of the cellulose–copper complex necessary for dissolution by cuene. All electron-irradiated fibers differed in swelling pattern from unexposed samples, but no dissolution of these fibers during observation was found. Of the neutron-irradiated samples, only those receiving the highest integrated flux, 10^{17} nvt, differed in swelling behavior from unexposed fibers. At this level no sausage formation was observed.

The difference in swelling of fibers receiving the largest quantities of gamma and of thermal neutron radiation is pictured in Figures 1 and 2. The swelling patterns of the unexposed controls are shown in Figures 1A and 2A respectively. The gamma-irradiated fibers show detachment of the primary wall, which has broken into small fragments and partially dissolved (Figure 1B), whereas neutron-irradiated fibers (Figure 2B) differ from the unexposed fiber only in generally reduced swelling and the lack of the sausagelike formations.

Swelling in Sodium Hydroxide and Absorption of a Direct Dye

Congo Red (C.I. 22120), a direct dye with a high substantivity for cotton cellulose, is routinely used to detect damage to the surface of cotton [10]. Fibers are first swollen in a 9% solution of sodium hydroxide. Immersion of the swollen fibers in aqueous solutions of Congo Red for a limited time results in the selective dyeing of the secondary-lamellae cellulose exposed at points of surface damage. For examination in the optical microscope, fibers are further swollen in 18% solutions of sodium hydroxide. Although more swelling ensues, new cracks and fissures are unstained.

Gamma-irradiated, unpurified fibers showed no dyeing of the secondary lamellae below doses of 10^7 r.; at higher doses, the fibers dissolved in the 9% alkali. Purified fibers showed slightly increased staining at 10^5 r., but no distinctive areas of damage were seen below doses of 10^7 r. At levels of 5×10^7 r. or more, the exposed, purified fibers also dissolved in 9% alkali. There were no differences in staining

of fibers irradiated in oxygen and nitrogen atmospheres. The electron-irradiated, unpurified samples showed little evidence of damage below dose levels of 10^7 r. Diffuse red areas were observed at this dose level, and samples receiving 10^8 r. dissolved in the 9% alkali. In neutron-irradiated fibers, evidence of moderate damage was first observed in samples exposed to 5×10^{16} nvt; those exposed to 10^{17} nvt stained red throughout the whole fiber.

Changes in the Fibrillar Structure of the Secondary Lamellae of Native Cotton

Fragmentation of Irradiated Fibers

When undegraded cotton fibers are beaten in water in a Waring Blendor for 20 min., the primary wall is detached from the fiber and the secondary lamellae are broken down into long sheets of fibrils, many of which are loose (Figure 3). This type of fibrillation is believed to be due to the fact that the lateral bonding forces of the fibrils (hydrogen bonds and van der Waals forces) are more easily broken than the valence forces in the cellulose molecules [25]. When previous degradation by any reagent has caused chain cleavage, the length and width of the sheets of fibrils decrease and short lengths of fibrils are more abundant in the specimen.

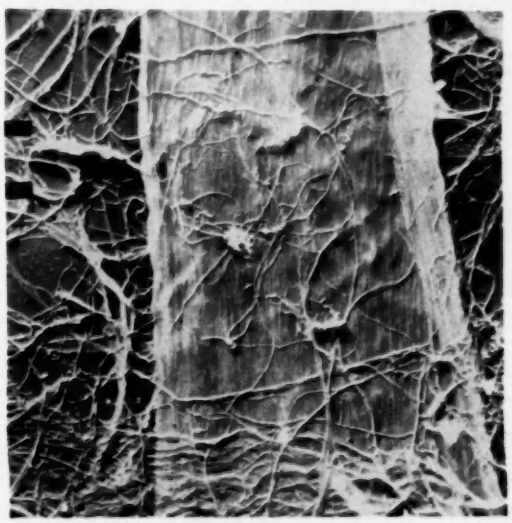
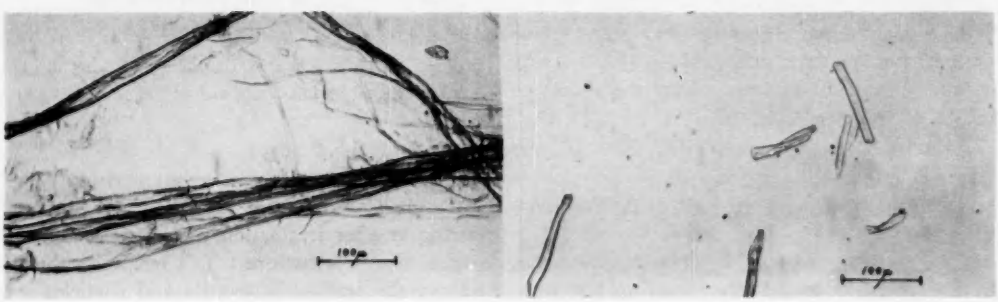


Fig. 3. Electron micrograph of fragment of unexposed purified cotton.

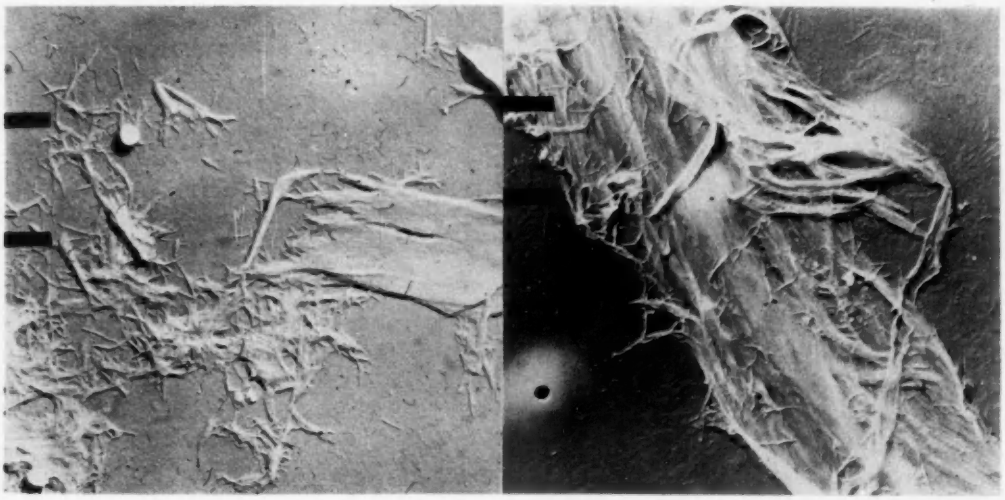
The progressive decrease in the size of fibrillar fragments of gamma-irradiated fibers with increasing dose is seen in the optical and electron micrographs in Figures 4A, 4B, and 5A. Distinctive changes in fibrillation pattern of either unpurified or purified fibers did not occur at levels of 10^5 and 10^6 r., but fibers receiving up to 10^7 r. formed shorter fragments, and at 10^8 r. some of the sheets of fibrils broke into short fragments and particles. The fragmentation pattern of the electron-irradiated fibers differed from that of unexposed fibers only at dose levels of 10^8 rep, but the same types of changes seen in gamma-irradiated fibers were found. The only changes noted in the patterns of neutron-irradiated samples were the formation of spicules, or loose ends of fibrils within the sheets of fibrils, and some evidence of shortening of the individual fibrils. These evidences of chain cleavage were not truly distinctive except in samples exposed to 10^{17} nvt (Figure 5B).

The effects of larger doses of gamma radiation on purified cotton are illustrated in Figure 6. The purified cotton was exposed in an air-dry condition to a dose of 4×10^8 r. in the Co^{60} source and then fragmented by the same technique used on the samples described above. Although excessive chain scission must have occurred to produce the small particles seen in most of the mircrographs, the predominant



A B

Fig. 4. Photomicrographs of fragments of gamma-irradiated purified cotton. (A) Unexposed; (B) 10^8 r.



A B

Fig. 5. Electron micrographs of fragments of irradiated purified cotton. (Compare with Figure 3.) (A) Gamma-irradiated, 10^8 r., N_2 atmosphere; (B) neutron-irradiated, 10^{17} nvt, in air.

change in pattern is in the formation of the short chunks of fibrils which appear fused and brittle. Most of the microfibrillate texture has been lost, and the smooth appearance suggests that interfibrillar fusion has occurred.

Irradiation of Fragments of Fibers and Fiber Cross Sections

Changes in fiber properties represent the integrated effect of action on the cellulose of the microfibrils in the secondary lamellae. More efficient and direct observations of changes in microfibrillar structure are possible when fibers are first beaten and then fragments of the secondary lamellae are reacted. Exposure of fragment specimens or of ultrathin (0.1 μ) fiber cross sections to the reagents under study have enabled more critical observation of the responses of fibrillar structures [25].

The micrographs in Figure 7 show the effects of irradiation of fragments of purified fibers. In Figures 7A and 7B, the total dose was 5.7×10^7 r.; the fragments in 7A were exposed in an air-dry condition, those in 7B were exposed while immersed in water and then dried. In Figures 7C and 7D, a dose of 1.5×10^8 r. was used. The fragments in 7C were exposed in an air-dry condition but were rinsed with several changes of water before the micrographs were made; the fragments in 7D were simultaneously exposed while suspended in water and dried before observing. Comparison of Figures 7A and 7C shows that reaction with water, either during or after irradiation, causes the disintegration of the fibrillar structure which results in the formation of the smaller fragments and particles seen in Figures 7B, C, and D, compared to the unchanged appearance of the fragments in Figure 7A.

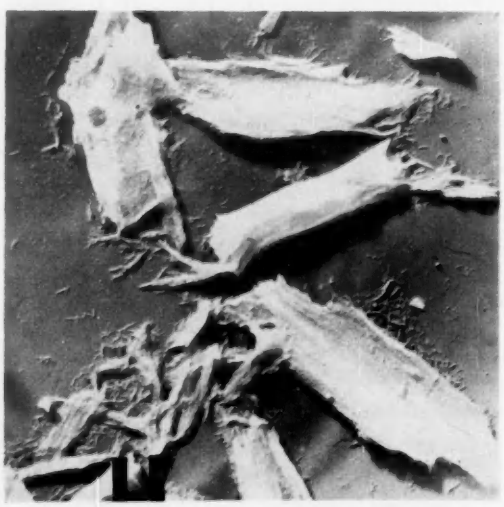


Fig. 6. Electron micrograph showing typical appearance of fragments of gamma-irradiated cotton, 4×10^8 r., in air.

The cross sections of unpurified fibers in Figure 8 were irradiated on the sample grids and received a total dose of 1.5×10^8 r. The section in Figure 8A, which was observed without further treatments, is no different in appearance from unirradiated sections. Following rinsing of the irradiated section with several changes of water (Figure 8B), most of the secondary wall cellulose is absent, although the primary wall appears intact. This apparent resistance of the primary wall probably results from the presence of the noncellulosic materials which are not converted into water-soluble products by the radiation.

Changes in Fibrillar Structure of Modified Cottons

When cottons are treated with reagents which modify the cellulose molecules by derivative formation or which polymerize within the interstices of the secondary lamellae, either with or without result-

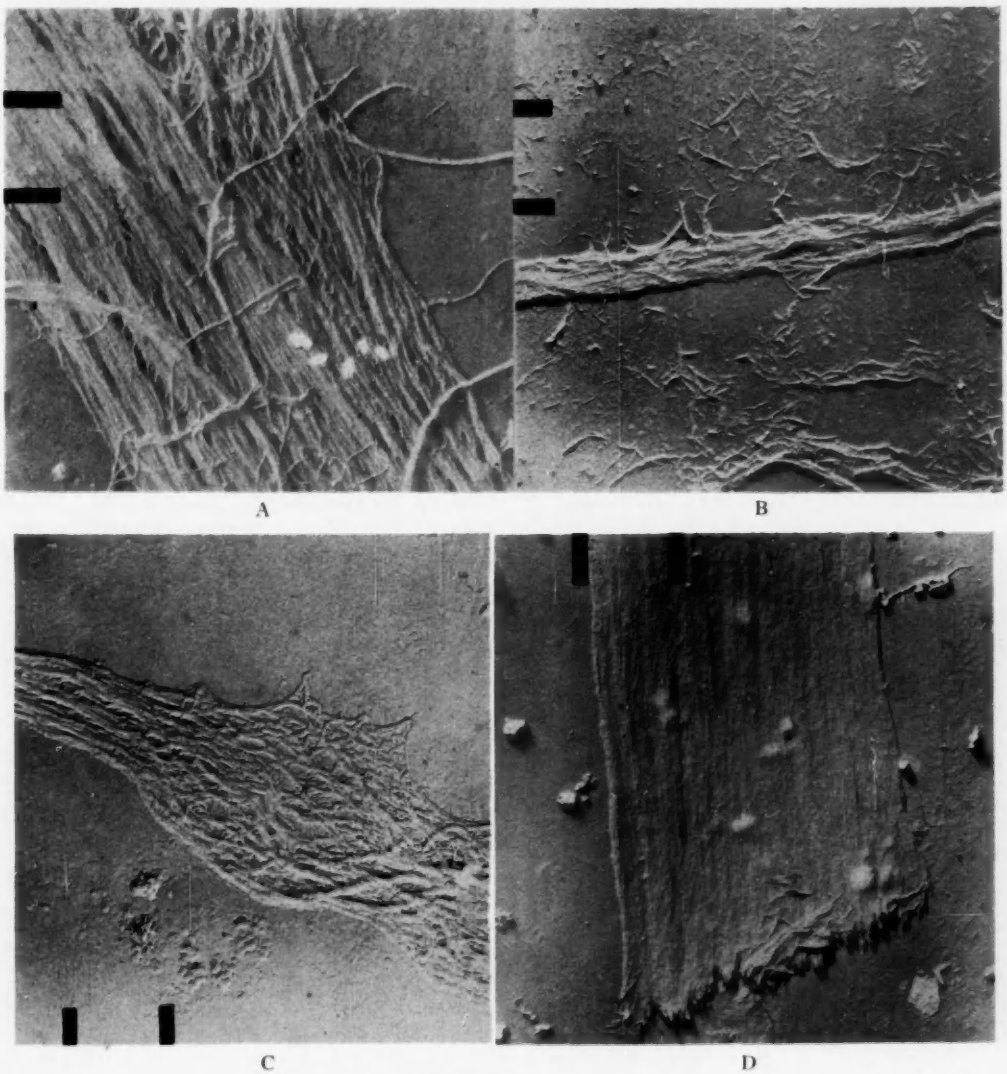


Fig. 7. Electron micrographs of gamma-irradiated cotton fragments. (A) 5.7×10^7 r., in air on specimen grid; (B) 5.7×10^7 r., in water; (C) 1.5×10^9 r., in air, rinsed with water; (D) 1.5×10^8 r., in water.

ant cross-linking of the cellulose chains, the lateral bonding forces of the chains are altered. This difference in lateral bonding can be demonstrated by the change in the fibrillation pattern resulting from beating in the Waring Blendor [25]. When interfibrillar bonding is increased, the fragments are shortened in length and fewer loose fibrils are obtained.

The effects of gamma radiation on the fibrillation pattern of the modified cottons listed below were evaluated by comparison of the pattern of unexposed cottons with those of the irradiated samples.

Partially acetylated [9] purified fabric (25% acetyl content)

Dichloropropanol-treated[2] purified fabric (1% weight gain)

Dimethylol cyclic ethyleneurea-treated[3] purified fabric (6% weight gain)

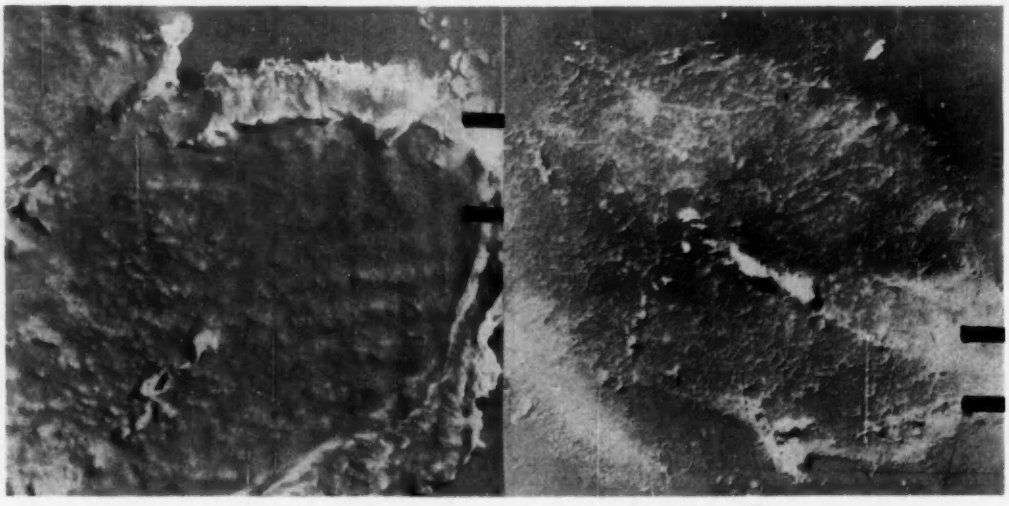
Acrylonitrile-treated[4] purified yarn (20% weight gain)

[2] Prepared using a mixture of 75% α,β- and 25% α,γ-glycerol dichlorohydrin by the method described by Reeves [18].

[3] Prepared using mixture containing 7% dimethylol ethyleneurea and 1.5% zinc nitrate catalyst. Sample dried at 60° C. for 7 min., cured at 160° C. for 4 min., washed once with soap, and rinsed twice with water [11].

[4] Yarns containing 80% pickup (by weight) of an aqueous solution of 0.4% potassium persulfate catalyst solution were exposed to acrylonitrile vapors at 80° C. for 10 min. [14].

Samples of the above cottons and a sample of untreated, purified fabric were simultaneously irradiated in an air-dry condition in the Co^{60} source, receiving a total dose of 5×10^7 r., and subsequently fragmented by the usual technique. There was very little change in the fibrillation pattern of the irradiated, partially acetylated sample except for the appearance of a few small particles. The fibrillation of the acrylonitrile-treated yarns did not differ from that of unmodified cotton in either the irradiated or unirradiated samples. However, in both samples there was some evidence of a smoothing or filming of the fibrillar surfaces which was not changed by the gamma radiation. Fragments of irradiated dichloropropanol-treated fabric were smaller than those found in the unexposed samples (Figures 9A and 9B), and the samples treated with dimethylol cyclic ethyleneurea were observed to be even more degraded following irradiation. The granulated appearance of the surface of the fibrils of the irradiated sample (Figure 9B) could be caused either by decomposition of the additive during irradiation or by extreme degradation of the cellulose. In the case of dichloropropanol-treated cotton, the weight increase was only 1% and the granular appearance probably was caused by more excessive degradation of the cellulose which had been exposed during treatment to a 25% solution of sodium hydroxide before the dichloropropanol was applied [18]. No pre-

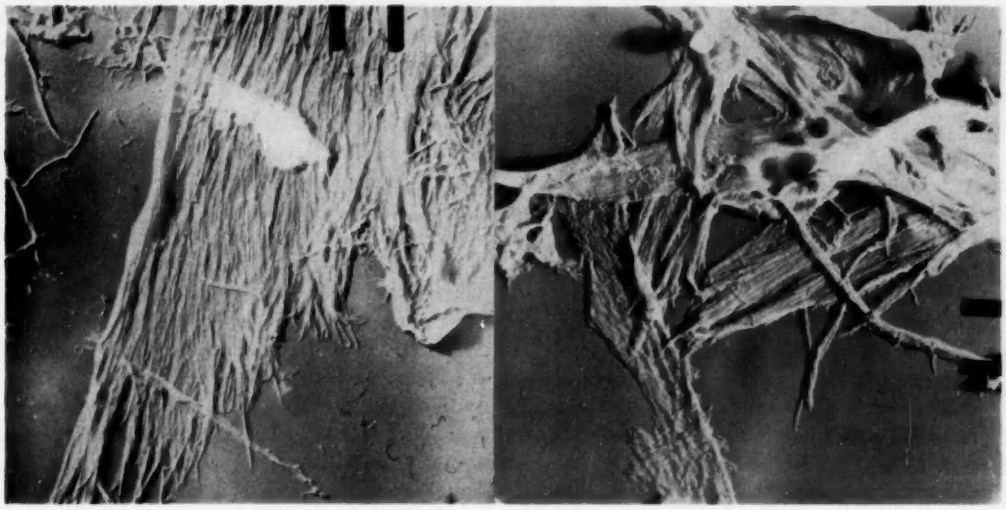


A B

Fig. 8. Electron micrographs of gamma-irradiated fiber cross sections, 1.5×10^8 r. in air. (A) Before rinsing with water; (B) after rinsing.

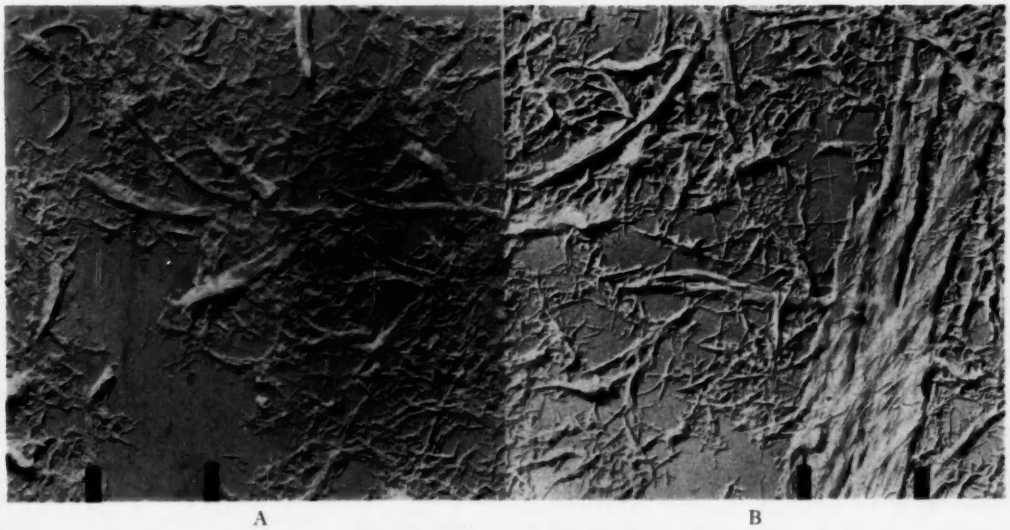
treatment of the prepared cotton was made before application of cyclic dimethylol ethylene urea; the similar granular appearance of the fibril surface after irradiation more likely was caused by disintegration of this reagent, which represents 20% of the exposed sample. Polymers of this type of reagent have been shown to be degraded by high-energy radiations [2], and the resulting irradiation products could cause further degradation of the cellulose.

Cottons with modified crystalline lattices were irradiated in the Co^{60} source, receiving total doses of 5×10^7 r. These modifications consisted of cellulose II, cellulose III [21], and amine-decrystallized cellulose I [20] structures. All samples having modi-



A B

Fig. 9. Electron micrographs of fragments of gamma-irradiated dichloropropanol-cotton reaction product. (A) Control; (B) 5×10^7 r., in air.



A B

Fig. 10. Electron micrographs of fragments of gamma-irradiated fiber, 0.5×10^8 r. (A) Cellulose II; (B) amine-decrystallized.

fied lattice structures appeared to be more degraded than samples with the original cellulose I structure. This is illustrated by the comparison of the micrographs in Figure 10 with those in Figure 4A of fragments of purified, unmodified cotton which had received a dose of 10^8 r.

Discussion

Effects of Radiation on Structure of Unmodified Cottons

One of the most sensitive microscopical tests used in these studies for detecting radiation damage is the staining of fibers with Nile Blue Sulfate and swelling in cupriethylenediamine hydroxide. Gamma rays, 2-Mev electrons, and thermal neutrons each caused sufficient difference in depth of staining, at the lowest doses used, to differentiate exposed from unexposed fibers. In the 0.2 *M* (in Cu) solutions of cupriethylenediamine, the swelling behavior of neutron-irradiated fibers could be differentiated from that of unexposed fibers only at the highest exposure, 10^{17} nvt, but swelling of fibers exposed to gamma rays or 2-Mev electrons differed from unirradiated samples at the lowest doses, 10^5 r. or rep. However, with increasing exposure, the sensitivity of these tests made differentiation between dose levels difficult.

The staining and swelling tests on gamma-irradiated, purified cotton agree qualitatively with the findings of Blouin and Arthur [3]. These investigators showed very little change in carbonyl and carboxyl group formation and in chain cleavages below doses of 5×16^6 r. in purified fibers irradiated in an oxygen atmosphere. At higher doses, there was a rapid increase in these parameters which, in the swelling test, was indicated by the increased solubility in the reagent. The somewhat smaller decrease in viscosity of electron-irradiated, unpurified yarns compared to values for dewaxed yarns, reported by Pan [16, 17], is reflected in the staining and swelling behavior of electron-irradiated, unpurified yarns and gamma-irradiated, unpurified fibers. The protection afforded by the noncellulosic materials probably involves complex radio-chemical phenomena.

Fragmentation of irradiated fibers followed by observation of the changes in fibrillation pattern in the electron microscope was a much less sensitive test of radiation damage than the optical methods used. However, it readily differentiated between doses in the range of 10^7 to 4×10^8 r. of gamma radiation or doses of 10^7 to 10^8 rep of 2-Mev electrons. Although the fiber is subjected to strong mechanical forces during beating, the only chemical and swelling reactions involve water; thus the degradation observed more nearly pictures the degree of chain scission which ensued during irradiation and does not show directly potential damage or group modifications on the cellulose chains. The smallest particles observed are approximately 1000 Å in length, or equivalent to a degree of polymerization of approximately 200. The predominance of the larger fragments observed in the micrographs suggests the presence of fibrils containing cellulose chains with a much higher degree of polymerization. However, the possibility exists that the length of the fragments obtained is not a simple function of chain scission. Blouin [3] reported a decrease in degree of polymerization of the gamma-irradiated samples from 4400 in unexposed samples to 800 at doses of 10^6 r., 200 at 10^7 r., and approximately 40 at 10^8 r.; values of intrinsic viscosity of electron-irradiated fibers reported by Pan [16] indicate similar decreases at equivalent doses. More extensive data are needed, however, before any correlation of fragment size and degree of polymerization is attempted.

Effects of Radiation on Some Modified Cottons

In studies of modified fibers, the fibrillation of irradiated acrylonitrile samples was not different from unexposed samples. This reagent can be polymerized by gamma irradiation [1] and has been shown to be more radiation-resistant than cellulose. Further tests are necessary to show whether or not the cotton cellulose is protected during irradiation. The material shown in the micrographs may be tubes of acrylonitrile only. The cross-linking agents, dimethylol cyclic ethyleneurea and dichloropropanol, failed to protect the cellulose from radiation damage, but this result was not unexpected. The amount of cellulose actually cross-linked is small and is believed to be confined to the poorly ordered regions [25]. Also, chain scission in cross-linked cottons at the β-1,4 glucosidic linkage may not necessarily be influenced by cross-linking between the cellulose molecules.

Formation of Particles Resembling Hydrocellulose

The presence of small fragments in micrographs of irradiated cottons resembling (but not positively

identified as) hydrocellulose particles suggests heterogeneity of the total reaction of cotton with radiation. The initial interaction of the radiation with atoms in the cellulose chains is probably homogeneous. The suggested heterogeneity is possibly caused by the greater reaction rates of induced secondary chemical reactions in the less ordered regions of the fiber, by differential accessibility of the regions to water (according to their original accessibility), by recrystallization, or by a combination of these causes. The micrographs of cottons with modified crystalline structure indicate increased degradation of the less crystalline cellulose compared to the degradation of unmodified cotton. Since no chemical or physical data were obtained on the irradiated samples of modified cottons, it is not known whether the decrease in fragment size results from increased chain scission or from the different response to swelling in water of these degraded modified cottons.

The Question of Damage by Thermal Neutrons

Micrographs of neutron-irradiated fibers after fibrillation show little evidence of damage at the exposures used. These results do not agree with the data reported by Teszler [23, 24]. However, the calculation of energy absorbed in a sample during exposure in a reactor generally includes contributions from fast neutrons, gamma rays, and secondary radiation from thermal-neutron capture reactions. For hydrogen, carbon, and oxygen, the capture cross-sections are so small that radiation from any radioactive isotopes formed contribute very little energy to the system. In the "very pure thermal neutron column" at the MTR, shielded by graphite and lead, the energy contribution from fast neutron and gamma rays is greatly decreased. Consequently, changes in physical and chemical properties of the cellulose should be less for the same thermal neutron exposure than in reactors where the fast neutrons and gamma rays are not as well shielded out [8]. Chemical data will be published showing that this is the case [5].

The damage seen in the micrographs of fibers exposed to an integrated flux of 10^{17} nvt of thermal neutrons could be caused by the small but measurable activity of the isotope formed from carbon, by activation of the container material, or by the cumulative gamma radiation.

Conclusions

Staining with Nile Blue Sulfate differentiates irradiated cotton fibers from unexposed cotton at the minimum dose levels observed (10^5 rep of gamma rays or 2-Mev electrons and 5×10^{14} nvt of thermal neutrons). Swelling behavior in 0.2 *M* (in Cu) cupriethylenediamine hydroxide solutions differentiates gamma- and electron-exposed fibers at the same dose level from unirradiated cotton, but no difference in the swelling behavior of control samples and thermal neutron-irradiated fibers occurs except after exposure to as high as 10^{17} nvt.

Fibrillation patterns of the secondary lamellae of irradiated, unmodified cottons are similar for each type of radiation investigated, in the range of irradiation studied, but thermal neutrons caused very little change in fibril dimensions.

Gamma irradiation, at dose levels of approximately 4×10^8 r., modified the fibrillation pattern of the secondary lamellae from a decrease in fragment size with increasing dose to the formation of larger, apparently solidified sections of material with altered fibrillar texture.

Small particles formed by fibrillation of fibers exposed to doses of 5×10^7 and 10^8 rep of gamma and electron radiation resembled, but were not identified with, hydrocellulose particles.

Acknowledgment

The authors wish to express their appreciation to Dr. Huo-Ping Pan and his associates, who furnished samples of electron-irradiated cotton; Miss Florine A. Blouin and Mr. Jett C. Arthur, Jr., Cotton Chemical Laboratory, who provided samples from cottons irradiated at MTR, Idaho Falls, Idaho for their studies and who performed all irradiations done in the Co^{60} facility at this laboratory; and to fellow members of Microscopy Investigations for assistance in making and processing electron micrographs.

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The Direct Nitration of Flax and Jute

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Abstract

Three samples of flax and one sample of jute were nitrated with a mixture of $HNO_3/H_3PO_4/P_2O_5$ for several lengths of time, ranging from ½ to 72 hr. The crude nitrate, Fraction A, was extracted with acetone and the acetone extract precipitated with water, giving three fractions: A1, the acetone-soluble fraction precipitated by water, A2, the acetone-insoluble fraction, and A3, the acetone- and water-soluble fractions. The DP's of Fraction A1 were determined. The mode of variation of the amount of Fraction A2 with time of nitration suggests the existence of intercarbohydrate links in jute but not in flax.

Introduction

In earlier studies [4, 5] it has been suggested that the method of direct nitration may be used to detect possible chemical links between cellulose and other carbohydrate components of naturally occurring cellulosic materials.

After nitration, the crude nitrate (Fraction A) may be separated into three fractions, A1, A2, and A3. Fraction A1 consists of nitrated alpha-cellulose, whereas Fraction A3 consists of nitrated hemicellulose. Fraction A2 contains both of these components. In wood, the yield of Fraction A1 increases

and that of Fraction A2 decreases with time of nitration. It was shown that these changes may be explained in terms of the breaking of a link between the alpha-cellulose and hemicellulose by the nitrating acid components.

This link is responsible for holding together the two components in Fraction A2. The method was applied to the wood of *Eucalyptus rostrata* (River Red Gum) and *Pinus halepensis* (Aleppo Pine). In the present paper the results obtained in the direct nitration of flax and jute are reported.

Experimental

Samples of flax were of the Concurrent variety grown in Beer-Tuvia, in the Southern region of Israel. Retting was carried out at 33° C. for 110 hr. The retted flax straw was dried in the open air and was then scutched. Green decortication was carried out on a flax scutching machine.

The 4-yr.-old retted sample was stored in an air-conditioned room at 20° C. and 70% RH. The jute fibers, taken from a lot of Pakistan raw jute, were extracted with alcohol:benzene (1:1) for 24 hr., dried, and carefully brushed to remove straw and tow.

The nitrating mixture was prepared according to Mitchell [7] and contained 64% HNO_3, 26% H_3PO_4, and 10% P_2O_5. It was made by adding the calculated amount of phosphorus pentoxide to the appropriate volume of 90% nitric acid. The addition was made slowly and the acid was kept at −16° C. and continuously stirred. After all the solid was added, the mixture was allowed to stand in the refrigerator, with occasional shaking, for 72 hr., after which it was filtered through glass wool and used immediately.

From each sample of fibers six 1-g. lots of the vacuum dried material were slowly added to 40 g. each of the nitration mixture at −16° C. The nitration was allowed to proceed in a refrigerator kept at 3° C. for various intervals of time ranging from ½ to 72 hr.

Washing of the nitration product was carried out according to Timell [11]. After the desired length of time the nitration mixtures were again cooled to −16° C. and the spent acids removed by filtration on a coarse sintered glass crucible. Care was taken not to allow air to pass through the nitrated samples at this stage. The nitrated samples were then washed successively with 50% acetic acid also cooled to −16° C., cold water, dilute acetic acid, and finally with large quantities of cold distilled water until the washings were no longer acid. The material was then extracted several times with methanol until the extract was colorless, dried in a vacuum, and weighed, this weight giving the yield of "crude nitrate" or Fraction A. The methanol treatment removes almost all the traces of lignin which were not dissolved by the nitrating acids. Fraction A was extracted several times with acetone until no more material could be extracted, the insoluble portion (Fraction A2) separated by centrifuging and the acetone solution poured into a large excess of distilled water, with continuous stirring, when fibrous, colorless cellulose nitrate (Fraction A1) was precipitated. To minimize degradation during the extraction stage, the air in the extraction flasks was replaced by nitrogen and the flasks were kept, as far as possible, in the dark. The precipitated nitrate was boiled for 5 min. with distilled water to remove all traces of acetone. All fractions were dried in a vacuum and weighed. Nitrogen contents were determined by a modified Kjeldahl method [2].

Degrees of polymerization (DP) were determined viscometrically in acetone. Corrections for nitrogen content were made according to Lindsley and Frank [6].

Results and Discussion

The results are given in Tables I–IV.

The individual results of the three flax samples show little dependence on the time of nitration. In particular, the yields of the fractions A, A1, and A3 are constant in each series. The values of the DP can also be regarded as constant. The very small value of Fraction A2 and the constancy of the yields of A1 and A3 show that in flax there is no link between the alpha-cellulose and the hemicellulose (= pectin) components.

The results for the three flax samples were averaged and collected in Table V. As is to be expected, the retted sample (III) has a higher alpha-cellulose content than the green decorticated, but four years of standing also reduces this value, as well as the DP.

The results for jute show a different picture. The variations in the yields of the various fractions show a similar behavior to that of wood [4]. Thus A and A2 decrease with time of nitration. A1 increases, reaching a maximum value which corresponds to the analytical alpha-cellulose content as determined by

TABLE I. Nitration of Flax, Sample I, Retted, 4 Years Old

Time, hr.	A, g.	A1, g.	A2, g.	A3, g.	Nitrogen, %	DP
2	1.34	1.19	0.01	0.14	13.4	4000
6	1.33	1.21	0.004	0.13	13.4	4100
18	1.28	1.16	—	0.12	13.5	3900
24	1.34	1.23	—	0.11	13.4	4000
48	1.31	1.20	—	0.11	13.6	3750
72	1.32	1.21	—	0.10	13.6	4000
Mean	1.32	1.20		0.12	13.5	3950

TABLE II. Nitration of Flax, Sample II—Green Decorticated

Time, hr.	A, g.	A1, g.	A2, g.	A3, g.	Nitrogen, %	DP
2	1.43	1.16	0.07	0.20	13.2	4000
5	1.43	1.17	0.05	0.21	13.3	4700
18	1.37	1.17	0.01	0.19	13.5	4850
24	1.36	1.16	0.01	0.19	13.3	4100
48	1.35	1.12	0.03	0.20	13.5	4200
72	1.37	1.12	0.03	0.22	13.6	4400
Mean	1.38	1.15		0.20	13.6	4400

TABLE III. Nitration of Flax Sample III—Retted

Time, hr.	A, g.	A1, g.	A2, g.	A3, g.	Nitrogen, %	DP
2	1.56	1.35	.03	0.18	13.3	4650
5	1.49	1.31	.02	0.16	—	
18	1.35	1.35	.01	0.19	—	
24	1.51	1.31	.01	0.19	13.6	
48	1.50	1.33	—	0.17	—	
72	1.50	1.33	—	0.17	13.6	
Mean	1.52	1.33		0.18		

the chlorite method. A3 increases, reaching a maximum, and then decreases. It was shown [5] that these phenomena may be explained in terms of an alpha-cellulose–hemicellulose link. The DP of the A1 fraction of the jute shows a sharp rise between ½ and 1 hr. of nitration, and a strong decrease between 24 and 72 hr. of nitration.

The results show good agreement with those reported by Timell [12] for the DP of the alpha-cellulose of jute and flax, which are 4700 for both, compared with the present study results of 4450 for jute and 4600 for flax. However, Timell obtained a maximum yield of alpha-cellulose from jute after 1 hr. of nitration, whereas in the present study the maximum yield is not reached even after 24 hr. of nitration (see Table IV). The difference is probably due to the difference in the temperature of nitration. Timell carried out the nitration at 17° C., whereas the present nitrations were carried out at 3° C. It was already shown [5] that the temperature of nitration greatly affects the yields of the various fractions and that the maximum yield of alpha-cellulose is reached sooner when the nitration is carried out at higher temperatures. Thus, in direct nitration of *Eucalyptus rostrata* wood, the yield of alpha-cellulose reached at 20° C. after 2 hr. of nitration is reached at 3° C. only after 120 hr.

The results obtained in the present study may be compared with previous work by other authors on the structure of flax and jute fibers. These works have been recently reviewed by Lewin [3]. Flax

TABLE IV. Nitration of Jute

hr.	A, g.	A1, g.	A2, g.	A3, g.	Nitrogen, %	DP	Alpha-cellulose
0.5	1.46	0.33	1.11		—	3950	—
1	1.45	0.33	1.09		—	4450	—
2	1.34	0.39	0.90	0.05	12.2	4250	23.8
10	1.32	0.48	0.62	0.22	13.3	4400	27.4
24	1.18	0.84	0.31	0.03	12.9	4200	49.0
72	1.12	1.06	0.03	0.03	13.5	1800	60.0
						(Anal.	61.1)

TABLE V. Comparison of Results for Three Flax Samples

Sample	A, g.	A1, g.	A3, g.	Nitrogen, %	DP	Alpha-cellulose	Alpha-cellulose (Anal.)
I Retted, 4 years old	1.32	1.20	0.12	13.5	3950	68	
II Green, decorticated	1.38	1.15	0.20	13.4	4550	65	
III Retted	1.52	1.33	0.18	13.5	4650	75	74.9

contains about 16% hemicellulose and 1.8–2.5% pectin. The former is contained in the outer middle lamella and is easily removed by means of dilute ammonium oxalate, whereas the latter appears in the inner middle lamella and is more difficult to extract. After isolation, both pectins show similar properties. It was suggested, therefore, by several authors [9] that pectin B is chemically linked to the cell wall. Stroink, Bendel, and Beerens consider these linkages to be "ester bridges" between the pectin and the cellulose of the cell wall. The results obtained in the present study indicate, however, that the cellulose in flax is not chemically linked to other carbohydrate components of the fibers. On the other hand, the existence of intercarbohydrate linkages in jute was suggested by Norman [8], but the work of Sarkar [10] and coworkers seems to indicate that such a linkage does not exist in jute. The present results, however, suggest the existence of such a linkage in jute.

It has often been suggested that in bast fibers the lignin is linked to the carbohydrate components [3] and that these links greatly affect the solubility of carbohydrate components. This is probably more the case in jute, which has a high lignin content (11.8%) as compared to flax (2.5%). These linkages are distinct from those described in the present paper since, during nitration, the lignin is partly oxidized and decomposed and partly forms nitrolignin. The oxidation products are dissolved by the nitrating acids, whereas the nitrolignin may be easily removed by methanol. The action of the nitrating acids on the lignin is comparatively rapid and is completed in the first few hours of nitration, while the first order reaction described in the present work continues up to 72 hr. of nitration.

The first order constant of the decomposition of the complex may be calculated from the linear plot of log A2 vs. time; the value found is 3.59×10^{-4}/min. This may be compared with 0.276×10^{-4} found for *Eucalyptus* at this temperature and 1.76×10^{-4} estimated for pine from the value at 20° C., assuming the ratio k_{20}/k_3 is the same for both woods. The plot of log A2 vs. time is shown in Figure 1.

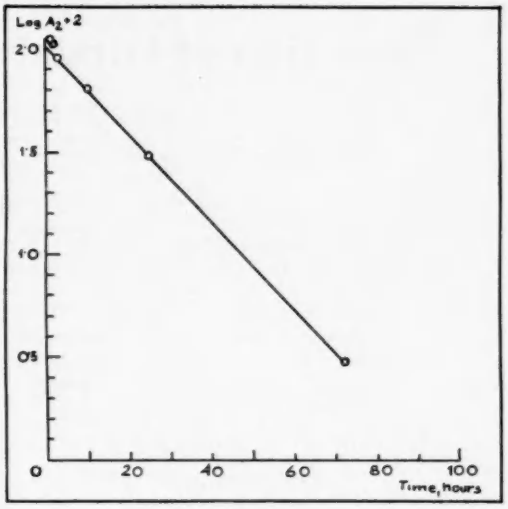

Fig. 1.

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Reaction of Dimethylol Urea with Cotton[1]

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Abstract

The reaction of dimethylol urea with cotton over a wide range of pH has been studied. Data are presented to show that the structure of the DMU polymer obtained with strongly acidic catalysts such as organic acids and ammonium salts is highly cross-linked, in contrast to the simple linear structure proposed by Steele and Giddings. Such a structure offers a reasonable explanation of the better washfastness of the DMU finish with these catalysts. The contribution of covalent cross-linking of the resin with cellulose in improving crease recovery is also discussed. The use of cuprammonium insolubility as an index of such cross-linking has been examined.

Introduction

Use of methylol ureas for improving crease recovery and dimensional stability of cotton and viscose fabrics is at least 25 years old. During this period, the effects of a number of process variables such as resin concentration, catalyst type and concentration, curing time and temperature, etc. on the properties of the treated fabric have been studied exhaustively and a great deal of literature is available on the subject. Little work has been published, however, on the chemistry of the polymerization of the "resin" within the fiber. Rumens [23] has studied the effect of molecular size and shape of the resin on its creaseproofing efficiency from theoretical considerations but does not give any experimental data for the polymer structure. Steele and Giddings [24] in a recent publication have studied the reaction of mono- and dimethylol urea with cotton and viscose, in the presence of ammonium chloride. From a chemical analysis of the product, they conclude that dimethylol urea polymerizes in the fiber to give an average DP of 2 and that this polymer is cross-linked to cellulose. They seem to imply that their conclusion holds true for DMU polymerized with all acid or potentially acid catalysts. On the other hand, the considerable published work [18, 25, 27, 28] on the structure of urea-formaldehyde resins shows that dimethylol urea, with its four functional groups, polymerizes extensively in the presence of acidic or potentially acid catalysts to form highly branched and cross-linked polymers. Furthermore, it is well known that the washfastness of DMU polymerized in cellulosic fibers varies considerably depending on the catalyst used. For DMU polymers of similar structures, this property should not vary to any considerable extent.

Another controversial aspect is the mechanism by which treatment with the resin improves crease recovery of the fabric. It has been shown [6, 11] that the crease recovery of the treated fabric is due to the improved elasticity of the fiber resulting from the resin treatment. Whether improvement in fiber elasticity is caused by the presence of a hard, amorphous resin, accompanied perhaps by hydrogen bond formation between the NH groups of the resin and the hydroxyl groups of cellulose, or whether it is due to a cross-linking of the hydroxyl groups of neighboring cellulose chains by the resin polymer through covalent bond formation is a subject of dispute. Cameron and Morton [5] were the first to suggest, from data on water imbibition, that at least part of the resin is involved in covalent cross-linking with cellulose. Lineken, Davis, and Jorgensen [15] studied the infrared spectra of cellophane treated with DMU and concluded that chemical reaction with cellulose was taking place. Gagliardi and Nuessle [9] suggest that crease recovery may be due to covalent cross links between cellulose and resin or to formation of strong hydrogen bonds. From analytical data of nitrogen and formaldehyde, insolubility in water, and high crease-proofing efficiency of the DMU polymerized in cotton, Steele and Giddings [24] conclude that the DMU polymer, which has an average DP of 2, must be cross-linked

[1] Part of the work reported here was presented at the Technological Conference held at ATIRA, Ahmedabad, October 1959.

with the cellulose. The ambiguous nature, at any rate for urea- and melamine-formaldehyde resins, of the arguments advanced to support cross-linking has been ably discussed by Nuessle [21]. Recently Marsh [17] has cautioned against the use of reduced water imbibition as evidence for cross-linking by referring to the work of Barret and Foulds [2] on the reaction of cellulose with formaldehyde which gives, under certain conditions, methylene bridge formation with high imbibition of water and aqueous solutions. It will be apparent from this review that, with available data, one cannot say conclusively that covalent cross-linking of cellulose is a *sine qua non* for crease recovery and that, at least in the case of polyfunctional reagents such as DMU, the alternative mechanism can equally well explain experimental facts.

The purpose of the present study was to collect information about the structure and DP of the polymer of DMU formed within the cotton fiber under different conditions of pH and to examine the possible cross-linking of the polymer with cellulose by studying the properties and behavior of the treated fabric.

Experimental Details

Materials

Fabric. The fabric used was a bleached, mercerized poplin (warp count 38s, weft count 38s, ends per inch 112, picks per inch 68), free from finish. This fabric was treated in boiling distilled water for half an hour, then washed with cold water three times followed by hydroextracting and air-drying. The pH of the fabric as determined by the method described later was 7.0.

Dimethylol urea was prepared as follows: 60 parts (1 mole) of BP urea and 170.3 parts (2.1 moles) of 37% formaldehyde were reacted for about 22 hr. at 25°–30° C. at a pH of 9.0 adjusted by suitable addition of sodium hydroxide. The resulting dense white product was filtered through a sintered glass funnel. The pH of the filtrate was 8.0, ensuring that the reaction had occurred throughout under only slightly alkaline conditions. The precipitates of DMU were washed thoroughly with distilled water, then with anhydrous ethyl alcohol, finally with diethyl ether, then dried in a vacuum desiccator.

Different melting points ranging from 126° C. to 140° C. have been reported in literature for DMU [25]. DMU prepared by the method given above had a melting point between 136° and 138° C. It was a white microcrystalline powder, soluble in water to the extent of about 15 g./100 ml. at room temperature, and showed a U:F molar ratio of 1:2. The dry DMU was stored in an airtight bottle in a refrigerator throughout the duration of the study and its stability was frequently checked by testing its melting point, solubility, and U:F ratio.

Catalysts. Hydrochloric acid, tartaric acid, ammonium sulfate, triethanolamine hydrochloride, magnesium chloride hexahydrate, and sodium bicarbonate were used as catalysts.

These were all of BP quality except hydrochloric acid, which was Analar. A wide range of pH from 3.5 to 9.5 was obtained for the cured fabric with these catalysts.

Application of DMU to Fabric

The air-dried and weighed sample of the fabric, 36 in. warpwise and 9 in. weftwise, was immersed for about 1 min. in a freshly prepared 10% solution of DMU containing the desired catalyst at its given concentration. It was then padded on a laboratory three-bowl mangle adjusted to give a wet pick-up of 75%. The actual wet pick-up was determined by weighing the padded sample and varied from 70 to 80%. After padding, the sample was immediately mounted on a metal frame with clips under just sufficient tension to remove wrinkles and dried on the frame for 8 min. at 110° C. in a thermostatically controlled forced draft hot air oven. The dried sample was cured on a metal pin-frame for 5 min. at 150° C. in the same oven. One fourth of the cured sample was cut out and kept apart for determining the pH of the cured fabric; the remainder was washed in 1% neutral soap solution at 80° C. for 15 min. (material:liquor 1:30) to remove catalyst residues, surface resin, etc. Washing was continued with distilled water till free formaldehyde was completely removed as tested by the color reaction with chromotropic acid salt as described later. The wet sample was hydroextracted, air-dried, and given a cold ironing to remove any wrinkles. With each catalyst, duplicate samples were padded and processed under identical conditions on different days. Samples were processed identically with distilled water and with DMU alone (without any catalyst). The water-treated sample was used as a control.

Testing

Where conditioning was necessary, the samples were conditioned at 80° F. and 65% RH at least overnight prior to testing.

pH of the aqueous extract of the fabric. Five grams of the dry fabric sample, cut in aproximately 1-cm. square pieces, was placed in a 500-ml. stoppered flask which had been previously rinsed with distilled water, 250 ml. of freshly boiled and cooled distilled water was added, and the sample was wetted by frequently shaking the flask for 1 hr. It was then left overnight; the pH of the supernatant extract was measured the next morning on a Beckman pH meter using a glass electrode.

Crease recovery was measured with the Monsanto Wrinkle Recovery Tester according to ASTM method D 1295-53T (1957). Ten readings each in warp and weft directions were taken for each sample. The sum of the mean warp and weft crease recovery is reported here.

Tear strength was measured with the Elmendorf Tear Tester according to ASTM method D 1424-56T (1957); four warpwise and four weftwise readings were taken for each sample. The sum of the mean warp and weft tear strength values in grams is reported here.

Nitrogen was determined by the macro Kjeldahl method, using potassium sulfate, copper sulfate, and sulfuric acid for digestion.

Total formaldehyde was determined by use of the chromotropic acid method described by Roff [22]. Approximately 0.5 g. of the treated fabric, accurately weighed (and given necessary moisture correction from a separate moisture determination) was hydrolyzed overnight in a stoppered 250-ml. flask with 100 ml. of 12 *N* sulfuric acid at room temperature. The contents were filtered through glass wool and 5 ml. of the filtrate was diluted to 100 ml. To 5 ml. of this, 1 ml. of 5% chromotropic acid sodium salt (BDH reagent for formaldehyde estimation) and 5 ml. of concentrated sulfuric acid were added and kept in a boiling-water bath for 30 min. After being cooled, this was made up to 100 ml. with distilled water and optical density of the solution was measured at 570 mμ by use of a photoelectric colorimeter. The formaldehyde content was calculated from a previously determined calibration curve for O.D. vs. formaldehyde content.

Methylol formaldehyde, that is, formaldehyde present as $N \cdot CH_2OR$ (R = H or glucose), was determined by the alkaline hydrolysis method of de Jong [13] described by Steele and Giddings, except that evaluation of formaldehyde was done colorimetrically using chromotropic acid as described before. Since acidification of an alkaline solution was involved here, all the reactants were previously cooled in ice to avoid any risk of loss of formaldehyde due to the heat of neutralization. Methylene formaldehyde, that is, formaldehyde present as $N \cdot CH_2 \cdot N$, was obtained as the difference of total formaldehyde and methylol formaldehyde.

Insolubility in Cuprammonium Hydroxide (CuAm)

Sufficient sample was weighed accurately so as to prepare exactly a 0.5% solution in cuprammonium hydroxide (15 g./l. copper, 200 g./l. ammonia) and placed in a centrifuge tube (vol. 30–35 ml.) which was then filled completely with CuAm. The sample weight was corrected for moisture, determined separately. Two small stainless steel balls (¼-in. diameter) were introduced for stirring and the tube closed with a rubber stopper; care was taken to exclude air. The centrifuge tubes containing the different fabric samples were stirred overnight at 6 rpm in a rotating box which protected them from light. Next day, the tubes were centrifuged for 10 min. at 2700 rpm in a laboratory centrifuge and the supernatant liquid was decanted off. The tubes were refilled with fresh solvent and the entire procedure of stirring, centrifuging, and decanting off was repeated. The residue in the centrifuge tubes was washed successively with fresh CuAm solution, with 28% ammonia solution, and then with 20% acetic acid till free from color. It was then filtered through a previously weighed sintered glass crucible No. 3, washed thoroughly with distilled water, anhydrous ethyl alcohol and ether successively, and finally dried in an oven at 110° C. for 3 hr. After being cooled and conditioned at 65% RH, the crucible with residue was weighed. (The moisture content of the residue was determined and necessary correction applied.) The reported value of CuAm insolubility is the weight of the dry residue expressed on 100 g. of bone dry fabric.

Results and Discussion

Table I gives the pH of the extract of the cured fabric, crease recovery, and tear strength of fabrics treated with DMU and different catalysts. Except

for the alkaline catalyst, which gives a slightly lower value, crease recovery does not vary much over the pH range used here for curing. These results are in conformity with the observation of Gagliardi and Nuessle [9] that over a pH range of 2 to 5 of the padding bath, crease recovery varied only from 73 to 78%. Hida [12] also found no significant effect of catalyst on crease recovery. The appreciable crease recovery obtained with sodium bicarbonate has been reported earlier by Gagliardi [8]. Murakami [19] commented that the alkaline catalysts used by Gagliardi were in effect latent acid catalysts due to the Cannizzaro reation between the catalyst and free formaldehyde always present during curing. Tear strength is generally related to crease recovery, except with the HCl catalyst, where there is distinct evidence of hydrolytic degradation of cellulose.

Table II gives values of residual formaldehyde, crease recovery, and CuAm insolubility of the samples after they were treated with 0.1 *N* HCl at 66° C. for 25 min. This treatment has been reported to hydrolyze the UF resin completely [3]. This was confirmed by nitrogen determination of the acid-hydrolyzed samples, which showed complete absence of N, indicating a complete removal of the resin. With all the catalysts, a small but significant amount of formaldehyde is retained by the cellulose even after the resin has been thoroughly removed. Lineken et al. [15], Wagner and Pacsu [26], and others have shown evidence for the formation of methylene cross links in the reaction of formaldehyde with cellulose under acidic conditions. That a similar reaction occurs during the curing process between cellulose and free formaldehyde evolved during curing has been proposed by Gagliardi and Nuessle [9] and experimentally demonstrated by Best-Gordon [4] and Alvsaker and Turner [1]. The latter authors also found, like the present authors, that even after removal of the resin by acid hydrolysis, some formaldehyde remained fixed and rendered the cellulose insoluble in CuAm. According to Best-Gordon, presence of 0.05% formaldehyde reacted with cellulose (forming methylene bridges) makes the latter insoluble in CuAm. Present data show that even with as little as 0.02% formaldehyde, the cellulose does not dissolve completely in CuAm. With the HCl catalyst, a significantly higher amount of residual formaldehyde is observed, which also gives to the fabric a small but significant crease recovery even after complete removal of the resin. With the alkaline catalyst, the situation is completely different. With as much as 0.04% residual formaldehyde, this sample is still completely soluble in CuAm. Perhaps, as Wood [29] believes, formaldehyde may react with cellulose under these conditions with the OH groups in the 2 and 3 positions of the same glucose units.

TABLE I. Properties of Fabrics Treated with DMU and Different Catalysts

Catalyst	Conc. of catalyst, % on wt. of DMU	pH	Crease recovery	Tear strength
None	—	7.3	166	1843
HCl	1.5	3.8	250	520
Tartaric acid	10.0	3.5	249	1139
$(NH_4)_2SO_4$	2.5	4.3	260	1017
T.E.A. HCl	10.0	4.9	248	1145
$MgCl_2$	30.0	6.3	252	1020
$NaHCO_3$	10.0	9.7	235	977
Distilled water	—	6.6	166	1843

TABLE II. Properties of DMU-Treated Fabrics After Acid Hydrolysis

Catalyst	HCHO, %*	Crease recovery	Cuprammonium insolubility
None	0.01	135	0.00
HCl	0.17	166	73.0–79.0
Tartaric acid	0.02	139	36.0–57.0
T.E.A. HCl	0.024	126	4.0–15.0
$(NH_4)_2SO_4$	0.087	134	62.0–81.0
$MgCl_2$	0.046	137	68.0–77.0
$NaHCO_3$	0.038	130	0.00

* Expressed on 100 g. of bone-dry fabric. Distilled water-treated sample gave 0.014% HCHO; formaldehyde values have been corrected for this blank value.

TABLE III. F:U Ratio for DMU Polymerized with Different Catalysts

Catalyst	Moles U	Moles F†	Moles F corrected	F/U ± S.E.*
None	0.038	0.035	0.035	0.91 ± 0.021
HCl	0.065	0.096	0.091	1.40 ± 0.012
Tartaric acid	0.061	0.091	0.091	1.49 ± 0.097
$(NH_4)_2SO_4$	0.065	0.095	0.093	1.42 ± 0.038
T.E.A. HCl	0.064	0.099	0.098	1.52 ± 0.023
$MgCl_2$	0.062	0.090	0.089	1.44 ± 0.044
$NaHCO_3$	0.055	0.075	0.074	1.33 ± 0.013

* Standard error = 2 × standard deviation.

† Distilled water-treated sample gave 0.003% HCHO; formaldehyde values have been corrected for this blank value.

TABLE IV. Ratio of Methylol and Methylene Formaldehyde in Fabrics Treated with DMU and Different Catalysts

Sample no.	Catalyst	Total corrected HCHO, %*	Formaldehyde as $N \cdot CH_2OR$, %*	Formaldehyde as $N \cdot CH_2N$, %*	$\frac{N \cdot CH_2N}{N \cdot CH_2OR}$
A_1	Nil	0.96	0.55	0.41	0.74
A_2	Nil	1.13	0.58	0.65	0.96
B_1	HCl	2.33	0.28	2.05	7.45
B_2	HCl	3.12	0.33	2.79	8.59
C_1	Tartaric acid	2.62	0.45	2.17	4.82
C_2	Tartaric acid	2.83	0.68	2.15	3.19
D_1	Amm. sulfate	2.85	0.50	2.35	4.71
D_2	Amm. sulfate	2.70	0.63	2.07	3.32
E_1	T.E.A. HCl	2.98	1.40	1.58	1.13
E_2	T.E.A. HCl	2.88	1.30	1.58	1.22
F_1	$MgCl_2$	2.65	1.48	1.17	0.80
F_2	$MgCl_2$	2.69	1.20	1.49	1.24
G_1	$NaHCO_3$	2.20	1.05	1.15	1.09
G_2	$NaHCO_3$	2.22	1.05	1.17	1.11

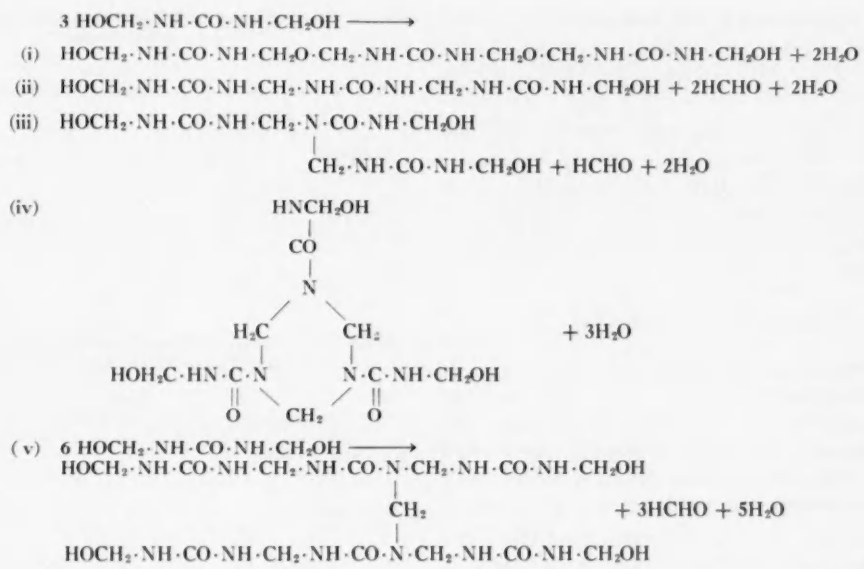
* Expressed on 100 g. of bone-dry fabric.

TABLE V. Comparison of Cyanimetric and Chromotropic Acid Methods for Formaldehyde Estimation

Sample no.	Mmoles of HCHO in 100-ml. solution determined by: Cyanimetric method	Chromotropic acid method
1	1.99	1.73
2	2.39	2.43
3	3.66	3.47
4	4.52	4.80
5	4.62	4.60
6	7.02	6.63

In Table III are given, for the different samples, formaldehyde : urea molar ratios, obtained from estimations of formaldehyde and nitrogen. The values of total formaldehyde were corrected for residual formaldehyde given in Table II to give only the formaldehyde coming from the resin. Each value of F/U is a mean of four readings obtained by duplicate analysis of two samples processed identically but on different days. The variation in N and formaldehyde within the sample was very small. Between samples, it was of a larger magnitude. Since formaldehyde varied proportionately with nitrogen ($r = 0.99$) and since the ratio F/U is the important parameter, it was considered more appropriate to calculate standard error for the ratios, rather than for N and formaldehyde values. Table III gives also standard error of the mean ratio F/U for each catalyst.

Table IV gives the individual values and ratio of methylene and methylol formaldehyde, defined and determined as described earlier. Duplicate determinations were carried out for each treatment. The variation in this case is much more than in Table III, probably because of the extrapolation involved. In spite of this, the difference in the value of the ratio for the different catalysts is of a much larger magnitude than the variation within a sample. Distinction among different catalysts is thus possible. Steele and Giddings have reported completely divergent results, their value for methylol formaldehyde being higher than that for methylene formaldehyde. Since a different method has been used in the present study for formaldehyde estimation, this was compared with the cyanimetric method used by Steele and Giddings. Six different formaldehyde solutions in 0.5 *N* NaOH were used for the comparison. The formaldehyde estimations by the two methods given in Table V show a very close similarity. The ratios $N \cdot CH_2 \cdot N / N \cdot CH_2OR$ given in Table IV are very well related with the acidity of the catalysts, the more acidic catalysts showing the higher ratios. Some of the possible modes of polymerization of DMU are shown in Figure 1. The calculated values of F/U and $N \cdot CH_2 \cdot N / N \cdot CH_2OR$ ratios for these structures are also shown. The change in the values of these ratios with increasing DP, branching, or cross-linking can be easily calculated. It will be apparent that no single structure can be assigned to the DMU polymer obtained with the different catalysts used here which would satisfy the analytical data given in Tables III and IV. In fact, a highly cross-linked polymer such as shown in Figure 2 has to be pos-



No.	F/U	Formaldehyde as $N \cdot CH_2O \cdot R$	Formaldehyde as $N \cdot CH_2N$	$N \cdot CH_2N$ / $N \cdot CH_2O \cdot R$
i	2.0	6	0	0
ii	1.33	2	2	1.0
iii	1.66	3	2	0.66
iv	2.0	3	3	1.0
v	1.5	4	5	1.25

Fig. 1. Possible Reactions During Polymerization of DMU

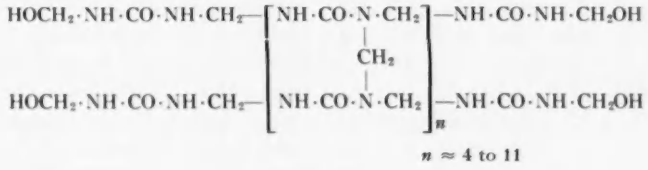


Fig. 2

tulated in the case of the strongly acidic catalysts. For the other three catalysts, a linear structure for the DMU polymer with a DP of three to six urea residues and probably with occasional branching in the case of the weakly acid catalysts satisfies the analytical data. These structures are more in conformity with observed facts than the simple structure proposed by Steele and Giddings for the DMU polymer irrespective of the catalysts used. In the first place, the known chemistry of urea formaldehyde resins, where strongly acidic catalysts are generally used, supports a highly cross-linked structure, as shown here. Furthermore, the proposed structures also offer a reasonable explanation for the differences in washfastness of DMU polymerized with different catalysts. Some typical data on washfastness are shown in Table VI. If the DMU polymer had the same simple linear structure proposed by Steele and Giddings irrespective of the catalyst used, it is difficult to explain the considerable variation in the washfastness of the finish obtained with different catalysts. Washfastness depends on the ease of hydroly-

sis of the polymer and of the methylene ether bonds between the polymer and cellulose in mildly alkaline conditions. For DMU polymers of similar structure, this property should not vary to any considerable extent. The greater DP, more complex structure, and considerable cross-linking within the resin polymer formed with strongly acidic catalysts explain the better washfastness obtained with these as compared with DMU polymerized with amine or metal salt catalysts.

Evidence for Cross-Linking

It has been mentioned in the introduction that one school of thought believes that a cross-linking of the cellulose with the resin polymer takes place with every resin–catalyst system used for the crease-resist finish and states, in fact, that such a cross-linking is an essential requirement for obtaining improved crease recovery. It has also been mentioned that most of the evidence in support of cross-linking is indirect, largely based on reduced water imbibition and insolubility in CuAm of the resin-treated cellulose. Where direct evidence is available, as with cyclic ethylene urea, the conclusion has been generalized to all types of resins and catalysts. Some of the criticism of these arguments for cross-linking has also been given in the introduction. Results of a preliminary examination of the CuAm insolubility test are given in Table VII to show its inconclusiveness as proof for cross-linking.

It can be seen that when the fabric is padded with DMU without a catalyst and dried at 110° C., it is completely soluble in CuAm, but on curing at 150° C., as much as 68% of the fabric remains insoluble. In the presence of tartaric acid catalyst, merely drying the fabric at 110° C. makes it practically completely insoluble; even after washing, its solubility is not increased significantly. Without a catalyst no cross-linking is possible. With tartaric acid catalyst, again, cross links cannot be present at the drying stage if one accepts the two-step mechanism proposed by Nickerson [20], Steele and Giddings [24], and others that polymer formation occurs during drying and cross-linking during curing when conditions are more favorable for ether formation. Again, no relation can be seen between crease recovery and CuAm insolubility. If cross-linking was responsible for both these properties, it may be expected that they should be related. Data in Table II show that fabrics treated with DMU and acidic catalysts show CuAm insolubility to different extents even after complete removal of the resin. Presence of methylene cross links formed by reaction between cellulose and free formaldehyde and accessibility of the test sample to the CuAm "solvent" probably play a significant part in determining the extent of CuAm insolubility.

TABLE VI. Effect of Different Catalysts on the Washfastness of UF Resin

Catalyst	Crease recovery, W + F			
	Before washing	After 1 wash	After 3 washes	After 5 washes
NH_4Cl	281	281	269	255
NH_4CNS	272	272	255	246
CAT.AC	260	253	235	205
$MgCl_2$	260	240	215	196
Tartaric acid	275	259	259	235

TABLE VII. Cuprammonium Insolubility of Cotton Fabric Given Different Treatments with DMU

Treatment	Cuprammonium insolubility, %	Crease recovery
DMU alone dried at 110° C.	0	133
DMU alone cured at 150° C.	67.9	166
DMU + tartaric acid dried at 110° C. before washing	99.2	220
DMU + tartaric acid dried at 110° C. after washing	95.5	189
Distilled water cured at 150° C.	0	166

The use of CuAm insolubility as an index for cross-linking between cellulose and resin suffers from an apparent contradiction in the particular case of DMU. Treatment with 0.5 *N* alkali has been shown to hydrolyze the methylene ether bonds between the —OH of cellulose and $—N\cdot CH_2OH$ of the resin. In fact, this is the method used for estimation of methylol formaldehyde. The reaction is practically complete in about 24 hr. One would expect a similar hydrolysis to occur during the CuAm insolubility test also, where the treated cellulose is in intimate contact with the highly alkaline reagent for 16–32 hr. Insolubility of the cellulose at the end of the test cannot then be due to cross links which do not exist any more.

In another experiment, fabric samples treated with

TABLE VIII. Alkaline Hydrolysis of DMU-Treated Cotton Fabrics

Catalyst	Time of hydrolysis, min.	Crease recovery	% HCHO
Tartaric acid	0	247	4.02
	20	236	3.405
	60	234	
	240	231	
	600	226	3.172
$MgCl_2$	0	248	
	15	218	
	30	212	
	60	208	
	240	199	

DMU in the presence of tartaric acid or magnesium chloride were treated with 0.5 *N* alkali at 20° C. (material : liquor ratio 1 : 200) and at periodic intervals were tested for crease recovery. The results are shown in Table VIII.

With the tartaric acid catalyst, there is a little loss of crease recovery, accompanied by a loss of 20.0% formaldehyde after 10 hr. of hydrolysis. The latter result indicates that hydrolysis of $N \cdot CH_2O \cdot Cell$ bonds is taking place. Thus, with this catalyst, the fabric retains most of its crease recovery even after any cross links that may be present have been hydrolyzed off. In the case of DMU polymerized with strongly acidic catalysts such as tartaric acid, a highly cross-linked three-dimensional structure has been suggested earlier for the polymer. Interstitial deposition of such a polymer in the accessible regions of cellulose, accompanied probably by hydrogen bond formation, appears to contribute most of the improved crease recovery to the fabric. If there is any cross-linking with cellulose, it seems to play a minor role in increasing fabric crease recovery. On the other hand, with magnesium chloride catalyst, there is a substantial loss in crease recovery under identical conditions of hydrolysis. Mere presence of the polymer, which in this case is essentially linear and has a low DP, is not enough to produce crease recovery. Cross-linking of the polymer with cellulose seems to be the essential requirement for crease recovery. In view of the high efficiency of magnesium chloride in promoting the etherification reaction between $N \cdot CH_2OH$ of the resin and OH of cellulose, shown by Frick et al. [7] for dimethylol ethylene urea, it is not unreasonable to assume that such a reaction should be favored in the case of DMU also.

Summary and Conclusions

The polymerization of DMU, when applied to cotton in the presence of catalysts with a wide range of pH, has been studied. From an analysis of N, $-N \cdot CH_2OH$, and $N \cdot CH_2 \cdot N$ in the treated fabrics it has been found that no one structure can be assigned to the DMU polymer formed with different catalysts to fit the analytical data. The polymer formed with strongly acidic catalysts such as tartaric acid and ammonium sulfate is highly cross-linked, whereas with the other catalysts it is essentially linear. Cross-linking of the polymer with cellulose is not a necessary requirement, in the former case, for producing crease recovery. The structures proposed here are in agreement with our present knowledge of the chemistry of urea-formaldehyde resins. They also offer a reasonable explanation of the wide difference in washfastness of the UF finish obtained with different catalysts.

Formaldehyde, liberated during drying and curing, reacts with the cellulose in the presence of acid catalysts and a small but significant amount of formaldehyde is detected in the fabric after complete removal of the resin by acid hydrolysis. This formaldehyde, probably present as methylene bridges, renders the acid-hydrolyzed fabric insoluble in cuprammonium hydroxide.

The use of insolubility in cuprammonium hydroxide of treated fabrics as a test for cross-linking has been examined and some preliminary data have been presented which question the validity of this test.

Acknowledgments

We are grateful to the Council of Administration of this laboratory for permission to publish these results. We are particularly indebted to Mr. D. V. N. Sarma for his help in the statistical analyses. One of us (R. D. M.) is grateful to the Government of India for the grant of a Fellowship.

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Reaction of Dimethylol Ethylene Urea with Cotton[1]

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Abstract

The reaction of dimethylol ethylene urea with cotton over a wide range of pH has been studied. It has been found that the DMEU polymer has a DP ranging from 2 to 5 depending on the acidity of the catalyst used. This variation in DP does not show a significant effect on fabric properties. The chlorine retention by DMEU-treated cotton fabrics has been reviewed and data have been presented to show the relation between this property and the methylol formaldehyde content of the treated fabric. A method has been suggested for estimating the methylol formaldehyde content. Data have also been presented examining the cuprammonium insolubility as a criterion for covalent cross-linking.

Introduction

With the growing importance of minimum care fabrics during the past few years, dimethylol ethylene urea (DMEU) has become one of the important resins in the textile finishing field. The application of this compound to cotton has been studied extensively by Cooke, Foster, Stam, and others [5, 6, 15]; the effect of various process variables such as resin concentration, type and concentration of catalyst, time and temperature of cure, etc. on fabric properties is well established. The chemistry of the polymerization of DMEU within the cellulosic fiber and of its possible reaction with cellulose has attracted attention only recently.

Cooke et al. [5] produced a polymer by heating monomeric ethylene urea formaldehyde at 130–135° C. without any accelerator for 4 hr. and ob-

[1] Part of the work reported here was presented at the Technological Conference held at ATIRA, Ahmedabad, October 1959.

tained a polymer with an average molecular weight of 2500, corresponding to a linear polymer approximately 17 units long. Gagliardi and Nuessle [9] postulated that the high creaseproofing efficiency of DMEU must be due to its cross-linking with cellulose, since the material does not form resins under the usual conditions of curing. Nuessle, Fineman, and Heiges [12] showed that, when a fabric finished with DMEU is mercerized without tension, there is a sharp reduction in its crease recovery without a significant reduction of the fixed resin content. This can happen only if the compound reacts with cellulose. Walter, Buxbaum, and Green [18] showed that DMEU reacting with itself in the presence of an acidic catalyst forms a glassy water-soluble polymer. On the other hand, when it reacts with celluluose or polyvinyl alcohol under similar conditions, it cannot be washed out with water. The reacted cellulose also becomes insoluble in cuprammonium hydroxide. On the basis of these results, they concluded that DMEU forms a linear polymer which is cross-linked with cellulose. Reid, Reinhardt, and Kullman [13] have suggested that, when polymerization of DMEU takes place, two types of linear polymers shown below are possible, depending upon the severity of curing.

Under mild conditions of curing a molecule of water is eliminated, with the formation of methylene ether bonds between two DMEU molecules. Under severe conditions of curing a molecule of formaldehyde is also liberated, along with a molecule of water, with the formation of methylene bonds. Using three catalysts (zinc nitrate, alkanolamine hydrochloride, and ammonium sulfate), Frick, Kottes, and Reid [7] showed from nitrogen and formaldehyde analysis that the polymerization of DMEU takes place with the liberation of formaldehyde and that the cross link formed with cellulose is about 1.4–1.5 ethylene urea residues in length.

Chlorine retention by DMEU-treated fabrics processed, stored, or laundered under certain conditions has received considerable attention recently. According to the mechanism accepted today, active chlorine from the laundering solution replaces a H atom in the >NH groups present in the resin polymer, forming chloramines which decompose at the high temperature of ironing, liberate HCl, and degrade the textile. Recent work has helped considerably in clarifying the puzzling situation that the DMEU-treated fabrics which theoretically cannot have any >NH groups should show chlorine retention.

The object of the present work was to study the polymerization of DMEU, applied to cotton, over a wide range of pH and to see if the changes in fabric properties obtained by the treatments could be understood in terms of the polymer structure.

Experimental Procedures

Materials

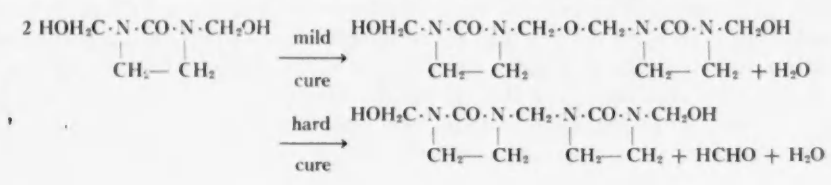
Fabric. The fabric used was 38s/38s cotton poplin (mercerized and bleached) with 112 ends and 68 picks/in. and was free from any finishing material. This fabric was boiled for ½ hr. with distilled water and washed till the washings gave a neutral pH. This step was necessary to remove any free alkalinity in the fabric.

Dimethylol ethylene urea. A typical commercial product was used. It contained 1.4% free formaldehyde, and gave a value of 1.74 for the ratio moles bound formaldehyde to moles CEU. The resin was stored in a cold storage room throughout this work.

Catalysts. To cover a wide range of pH and to include all types of catalysts commonly used in industry, the following catalysts were selected: hydrochloric acid, tartaric acid, ammonium sulfate, triethanolamine hydrochloride, magnesium chloride hexahydrate, and sodium bicarbonate. These chemicals were all of B.P. quality except HCl, which was Analar.

Application of DMEU

The padding bath was prepared by diluting the liquid resin by addition of distilled water, adding predetermined concentration of catalyst, and diluting



to required volume. 36 × 10 in. pieces of the fabric were cut, the longer side being the warp, and padded on a three-bowl laboratory padding mangle adjusted to give a wet pick-up of 75%. Wet pick-up varied from 70 to 80%. The padded sample was clamped to its original width on a frame of the same size and dried in a forced draft thermostatically controlled hot air oven at 110° ±1° C. for 8 min. The sample was then loosely suspended on a pin frame and was cured at 150° C. for 5 min. After the curing was over, the fabric was washed at 80° C. in a solution containing 1.0% neutral soap for 15 min. using a material : liquor ratio of 1 : 30. The sample was then washed with water till the washings gave a neutral pH test and no positive test for formaldehyde with chromotropic acid (sodium salt).

Samples were also padded with DMEU alone (no catalyst) and with distilled water and processed as above. The distilled water-treated sample was used as a control.

All samples were prepared in duplicate by repeating the entire experiment on a different day.

Testing Methods

pH of the cured fabric, crease recovery, tear strength, nitrogen and total formaldehyde content, and insolubility in cuprammonium hydroxide (CuAm) of the various fabric samples were measured according to the procedures described earlier [10].

Loss of tensile strength due to chlorine retention was determined according to AATCC Tentative Test Method 92-1958, using a 3-in. test length on the Instron Tensile Tester for tensile strength measurements.

Results and Discussions

Table I gives pH of the cured fabric, resin fixation (expressed in terms of N content), crease recovery, and tear strength of fabrics treated with DMEU and different catalysts. It is interesting to note that, even without any catalyst, as much as about 3% resin is fixed in the fabric, without any improvement in crease recovery. With the alkaline catalysts, only about 0.6% resin has been fixed, in contrast to dimethylol urea, where substantial resin fixation and crease recovery have been observed using alkaline catalysts [8, 10]. In view of the poor resin fixation obtained with the alkaline catalyst, it was excluded from further studies. The five acidic (or latent acid) catalysts have given very similar values of resin fixation and crease recovery, except tartaric acid, which gave a somewhat lower crease recovery. Tear strength also does not show any pronounced differences, except in the case of HCl catalyst, where there is distinct evidence of a hydrolytic degradation of the cellulose.

The evolution of free formaldehyde during drying and curing of creaseproofing resins has been observed by many workers. Gagliardi and Nuessle [9] pointed out the possibility that, under the usual conditions of curing, this free formaldehyde could react with cellulose to form methylene bridges. This has been experimentally confirmed for UF resins by Alvsaker and Turner [1] and also in an earlier publication from this laboratory [10]. Steele [16], Wagner and Pacsu [17], and others have shown that formaldehyde, reacting with cellulose under acidic conditions, forms methylene bridges between neighboring cellulose molecules. To examine such a possibility for the DMEU-treated fabrics, they were hydrolyzed in 0.1 *N* HCl at 66° C. for 40 min. Cooke, Dusenbury, Kienle, and Lineken [4] have shown that, under such mild conditions of acid hydrolysis, formaldehyde reacted with cellulose cannot be removed. Much more drastic hydrolysis, sufficient to degrade the cellulose highly, is necessary to remove such formaldehyde. Complete removal of the resin in the acid-hydrolyzed samples was checked by nitrogen determination. Formaldehyde, present as methylene bridges, was determined by treating the acid-hydrolyzed fabric sample in 12 *N* H_2SO_4 overnight at room temperature in a tightly stoppered Erlenmayer flask, followed by a colorimetric evaluation of formaldehyde using chromotropic acid salt. It had been established previously that longer times of treatment with 12 *N* H_2SO_4 did not give significantly higher values for formaldehyde. Table II gives the results of formaldehyde content, crease recovery, and CuAm insolubility of the DMEU-treated fabrics after acid hydrolysis. With each catalyst, a small but significant quantity of residual formaldehyde is detected after complete removal of the resin. Except the HCl catalyst, where this quantity is appreciably high, residual formaldehyde content varies between 0.02 and 0.05% on the weight of the fabric. The same range of formaldehyde content has been reported by Best-Gordon [3] in a number of commercial samples examined by him.

TABLE I. Properties of Fabrics Treated with DMEU and Different Catalysts

Catalyst	Concentration of catalyst, % on wt. of resin solids	pH of cured fabric	Nitrogen content, %*	Crease recovery	Tear strength
None	—	6.9	0.59	152	2122
HCl	1.5	3.5	1.64	262	426
Tartaric acid	10.0	3.4	1.49	246	1140
$(NH_4)_2SO_4$	2.5	5.1	1.34	272	1092
T.E.A. HCl†	10.0	4.7	1.43	278	1117
$MgCl_2 \cdot 6H_2O$	30.0	6.6	1.31	272	968
Sodium bicarbonate	10.0	8.9	0.12	148	1862
Water	—	6.7	0.00	141	1755

* Expressed on 100 g. of bone-dry fabric.
† Triethanolamine hydrochloride.

CuAm insolubility shows a good agreement with residual formaldehyde content. In the case of the HCl catalyst, the high concentration of methylene formaldehyde has produced a small but significant improvement in crease recovery. These results are in close agreement with those reported earlier for DMU-treated cotton [10]. This may be expected since, irrespective of the source of formaldehyde, its reaction with cellulose in the presence of different catalysts should be the same.

Results of total N, expressed as moles of CEU (cyclic ethylene urea) and of total formaldehyde as moles are given in Table III. Since the concentration of formaldehyde present in the resin polymer (and not total formaldehyde) is needed for the consideration of the structure of the latter, values for total formaldehyde have been corrected for formaldehyde present in the form of methylene bridges (Table II); the corrected values are given in Column 4 of Table III. The ratio moles F to moles CEU given in Column 5 has been obtained as the mean of four values obtained independently on two duplicate samples processed on different days. The standard error of the mean has also been given as twice the standard deviation.

TABLE II. Properties of DMEU-Treated Fabrics after Acid Hydrolysis

Catalyst	Formaldehyde, %*	Crease recovery	Cuprammonium insolubility
None	0.0048	158	00
HCl	0.2210	178	80
Tartaric acid	0.0189	139	42
$(NH_4)_2SO_4$	0.0212	140	40
T.E.A. HCl	0.0288	151	67
$MgCl_2 \cdot 6H_2O$	0.0469	152	82

* Expressed on 100 g. of bone-dry fabric. Water-treated sample gave 0.0213% formaldehyde; this correction has been applied.

Having two functional groups, DMEU can form only linear polymers, unlike DMU. It is not necessary, therefore, in the present case to differentiate formaldehyde present as $>N \cdot CH_2OR$ and as $>N \cdot CH_2 \cdot N<$ in order to ascribe a structure to the DMEU polymer within the fiber. This is a fortunate circumstance, since it will be seen later that the methylene ether bond between the terminal

TABLE III. F/CEU Ratios for DMEU Polymerized with Different Catalysts

Catalyst	C.E.U., mole	Formaldehyde, mole*	Formaldehyde, corrected, mole	F/CEU ± standard error†
None	0.021	0.0309	0.0309	1.46 ± 0.036
HCl	0.059	0.0793	0.0719	1.23 ± 0.016
Tartaric acid	0.053	0.0673	0.0666	1.26 ± 0.045
$(NH_4)_2SO_4$	0.048	0.0735	0.0728	1.53 ± 0.047
T.E.A. HCl	0.051	0.0774	0.0765	1.51 ± 0.031
$MgCl_2 \cdot 6H_2O$	0.047	0.0666	0.0651	1.39 ± 0.059

* Water-treated sample gave 0.003% formaldehyde; this correction has been applied.
† Standard error = 2 × standard deviation.

$>N\cdot CH_2\cdot OH$ groups in the DMEU polymer and $-OH$ groups of cellulose is not alkali-labile as in the case of the DMU polymer.

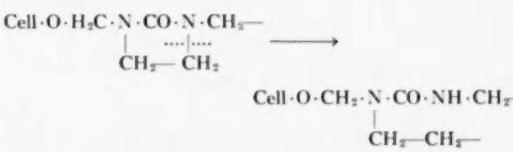
From the results given in Table III an average DP varying from about 2 to 5 can be calculated for the DMEU polymer formed with the different catalysts. Frick, Kottes, and Reid [7] found a value of the ratio F/CEU ranging from 1.4 to 1.7 with ammonium salt, alkanolamine, and metal salt catalysts. Since maximum cross-linking of cellulose with the DMEU polymer occurs with the metal salt catalysts, as seen from their other data, they assign an average DP of 1.4 to 1.5 to the DMEU polymer based on the value 1.7 for F/CEU obtained with these catalysts. With the other two catalysts, they assume a loss of formaldehyde by decomposition, under curing conditions, of free $N\cdot CH_2OH$ groups left unreacted with cellulose, due to the lower cross-linking efficiency of these catalysts, giving lower values of F/CEU. It will be seen from subsequent data presented here that free $N\cdot CH_2OH$ groups can withstand curing conditions and do exist in the cured fabric. Taking this into account, the results for DP agree substantially with those proposed by Frick et al. It is interesting to note that, with the two strongly acidic catalysts, the DP has increased by a factor of 2 to 3 without producing a significant difference in fabric properties.

A critical examination of the CuAm insolubility test as a criterion of cross-linking has been given earlier [10]. Some results with DMEU are shown in Table IV. In the case of DMEU, it has been established reasonably beyond doubt that crease recovery is due to cross-linking. If CuAm insolubility was also a result of cross-linking alone, a simple relation between the former property and crease recovery may be expected. The data in Table IV show that this is not true. In fact, most of the samples show little improvement in crease recovery over the water-treated control and yet are insoluble in CuAm to the extent of 80% or more. Furthermore, even under processing conditions which are very unlikely to form either methylene bridges or cross links of the DMEU polymer with cellulose, the fabric sample is still insoluble in CuAm. For example, cross-linking is not possible in the treatment without any catalyst, and improbable in overnight drying at room temperature with either tartaric acid or magnesium chloride catalysts. Yet, all these three samples show a CuAm insolubility exceeding 90%. It is clear that cross-linking of the cellulose is not the only factor contributing to insolubility in CuAm.

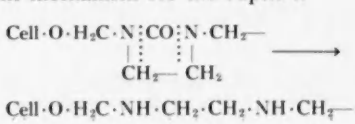
Chlorine Retention

According to the accepted mechanism of chlorine retention, DMEU-treated fabrics should not suffer from this drawback. In practice, however, this has not been found to be true. Damage due to retained chlorine has been observed generally under the following circumstances: use of ammonium salt or alkanolamine hydrochloride catalysts; omitting the process wash which should be given to the cured fabric, thus leaving catalyst residues in the fabric; including an acid souring step in laundering; and mild curing with metal salt catalysts. Chlorine retention may be observed immediately after the resin treatment or may develop gradually during successive alkaline washes. The importance of testing for chlorine retention after repeated washing was recognized by Bacon, Smith, and Hughes [2] who found that curing with $MgCl_2$ or zinc nitrate catalysts at 180° and 160° C. respectively gave practically complete freedom from chlorine retention even after 10 launderings.

During the last year, attempts have been made to explain these experimental observations. A brief review of this work may be useful before discussing our data. Mosher [11] states that, during the laundering of DMEU-treated fabrics, the CEU ring may be ruptured as shown below, forming sites for chlorine retention.



He ascribes the initially low chlorine retention observed with metal salt catalysts to the buffering action of the intrafiber deposition of metal oxides and carbonates. With repeated laundering, these will be progressively washed out, leading to increasing chlorine damage. Frick et al. [7] also consider rupture of the CEU ring structure responsible for chlorine retention. They, however, have proposed a different mechanism for the rupture.



Differences in initial chlorine retention behavior due to catalyst type are, according to these authors, because of the different cross-linking efficiencies of catalysts. Reacting

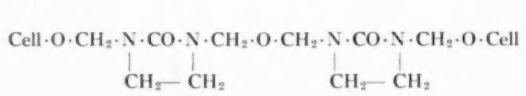
$$\begin{array}{l} CH_2 \cdot N \cdot CO \cdot N \cdot CH_2OH \\ \quad | \qquad\quad | \\ CH_2 - \quad CH_2 \end{array}$$

with cellulose in the presence of different catalysts and using percent nitrogen loss on washing as an index of the extent of reaction with cellulose, they rank zinc nitrate, alkanolamine hydrochloride, and ammonium salt catalyst in decreasing order of efficiency for promoting the reaction between the terminal $>N \cdot CH_2OH$ group of the reagent with the $-OH$ of cellulose. Since reaction of DMEU with cellulose is essentially similar, they consider this also as the order of cross-linking efficiency of the catalysts. Thus, with a catalyst having poor cross-linking efficiency, a large number of polymer chains may have reacted with cellulose at only one end or not at all, leaving free $>N \cdot CH_2OH$ groups. They claim that under usual curing conditions these free methylol groups cannot exist; they decompose to give $>NH$ with liberation of formaldehyde. The poorer the cross-linking efficiency or milder the curing, the greater the number of sites for chlorine retention.

Smith [14] considers it very unlikely that a stable structure such as CEU can be ruptured by a mild alkaline treatment as given in laundering. He has proposed an entirely different mechanism which, to the present authors, appears more plausible. According to Smith, the cure is incomplete with ammonium salt or organic acid catalysts; some of the DMEU molecules may have reacted with only one molecule of cellulose, leaving free $>N \cdot CH_2OH$ groups. During an alkaline treatment, these would lose formaldehyde and form $>NH$ groups, which are the sites for chlorine retention. Such fabrics would thus be initially chlorine retentive. On the other hand, all DMEU-treated fabrics irrespective of catalyst or curing conditions used show chlorine retention on storage if they are not washed off after curing. This is due to an acid hydrolysis of the cross link between cellulose and DMEU due to the catalyst residues. Again, $>N \cdot CH_2OH$ groups are formed which decompose to $>NH$ during alkaline treatments. If fabrics treated with DMEU and metal salt catalysts under mild curing conditions are stored and given an alkaline wash, they again show a strength loss of 20–40% due to chlorine retention, even if they have been given a wash-off after curing. Smith suggests that, under these conditions, DMEU could polymerize and react with cellulose to give

TABLE IV. Cuprammonium Insolubility and Crease Recovery of Samples Treated with DMEU under Different Conditions

Treatment	Cuprammonium insolubility	Crease recovery
DMEU alone—dried at 110° C; 8 min.	99.0	152
cured at 150° C; 5 min.		
dried at 110° C., 8 min.	00.0	152
DMEU + Tartaric acid—dried at 110° C., 8 min.	96.0	226
—dried overnight at room temperature	94.0	180
—dried at 110° C., 8 min., then washed	97.0	180
DMEU + $MgCl_2 \cdot 6H_2O$—dried at 110° C., 8 min.	89.0	168
—dried overnight at room temperature	91.0	127
—dried at 110° C., 8 min., then washed	84.0	135
Water	00.0	141



Such a possibility has been suggested earlier by Reid et al. The ether linkage between two nitrogen atoms would be more susceptible to hydrolysis than that between a nitrogen and cellulose. Such a hydrolysis would again give $>N\cdot CH_2OH$ groups, which cause chlorine retention, as seen earlier.

If this is the mechanism by which chlorine retentive sites are formed, it is obvious that chlorine damage should be related to the number of $>N\cdot CH_2OH$ groups either present initially in the finished fabric or developed during storage or laundering. An attempt was made to determine $>N\cdot CH_2\cdot OH$ in DMEU-treated fabrics using different conditions of alkaline hydrolysis followed by colorimetric estimation of formaldehyde by the chromotropic acid method. It was found that hydrolysis with 0.5 *N* sodium hydroxide at 35° C. for 24–28 hr. was suitable for the determination. Crease recovery and chlorine retention of the samples were measured before and after the hydrolysis; results are given in Table V. It is obvious from the crease recovery data that only free NCH_2OH is being estimated and that the ether linkages formed between the DMEU polymer and cellulose are not affected at all. Frick et al. [7] have also observed that alkaline hydrolysis, which distinguishes between methylene ether and methylene linkages in DMU-treated cellulose, fails to give similar information in the case of DMEU.

Data given here do not support the statement of Frick et al. that free methylol groups cannot exist under curing conditions. They are present to varying extents depending on catalyst type and can be readily estimated.

Table V also shows that the strength loss due to chlorine retention and scorching is directly related to the free methylol content in the cured fabric. The data support the observation by Frick et al. that metal salt catalysts are the most efficient and ammonium salts (and organic acids) the least efficient catalysts for promoting cross-linking. The alkanolamine catalyst has, however, given much better results here than observed by them. With the magnesium chloride catalyst, increasing the severity of curing has reduced the potential chlorine retention damage accompanied by a reduction also in free $>N\cdot CH_2OH$ content. The chlorine damage before alkaline hydrolysis is due to a partial decomposition of $>NCH_2OH$ to $>NH$ either during curing as suggested by Frick et al. or during the treatment with hypochlorite at pH 9.5 as specified in the standard test for chlorine retention. The strength loss due to retained chlorine after alkaline hydrolysis shows the potential degradation that the fabric would suffer on repeated washing. These results again agree well with the methylol content in the cured fabrics. Further work on the problem is in progress and will be reported later.

Summary and Conclusions

When DMEU is applied to cotton in the presence of acidic or latent acid catalysts, the DMEU polymer formed has a DP of 2 to 5, the higher DP being obtained with the more acidic catalysts. No marked differences were observed in crease recovery with any of these catalysts. Alkaline catalysts gave poor

TABLE V. Alkaline Hydrolysis of Fabrics Treated with DMEU and Different Catalysts

Catalysts	% Formaldehyde evolved after hydrolysis for			% loss in breaking strength due to scorching		Crease recovery after 28 hr. of hydrolysis	
	28 hr.	76 hr.	125 hr.	Before hydrolysis	After hydrolysis	Before	After
None	0.177	0.177	0.185	86	92	—	—
HCl	0.062	0.076	0.076	6	22	230	228
Tartaric acid	0.171	0.171	0.171	54	74	214	220
$(NH_4)_2SO_4$	0.145	0.145	0.145	58	68	254	252
T.E.A. HCl	0.038	0.047	0.047	0.7	9	271	267
$MgCl_2\cdot 6H_2O$	0.069	0.081	0.078	0.8	10	278	280
$MgCl_2\cdot 6H_2O$, hard cure*	0.028	0.040	0.042	0.9	1	278	278
Water	0.000	0.000	0.000	0.0	—	—	—

* Cured at 175° C. for 5 min.

resin fixation and crease recovery, indicating little or no polymerization and reaction with cellulose. In absence of any catalyst, a significant amount of fixed resin was obtained without improvement in crease recovery.

Formaldehyde liberated during drying and curing reacts with the cellulose and is detected in the fabrics after complete removal of the resin by acid hydrolysis. This formaldehyde, probably present as methylene bridges, renders the acid-hydrolyzed fabric insoluble in cuprammonium hydroxide.

Alkaline hydrolysis of the DMEU-treated fabrics with 0.5 *N* NaOH at 35° C. gives an estimate of free methylol groups present in the cured fabric and which form chlorine retention sites either initially or after repeated laundering. The methylene ether bonds between the DMEU polymer and cellulose are stable to this alkaline hydrolysis. Methylol content determined in this manner shows a good relation with strength loss due to chlorine retention and scorching.

Acknowledgments

We are grateful to the Council of Administration of this laboratory for permission to publish these results. We are particularly indebted to Mr. D. V. N. Sarma for his help in the statistical analyses.

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Letters to the Editor

Interpretation of Staining Results Obtained by the Procion Black HGS Procedure

Central Research Laboratories
Toyo Rayon Co. Ltd.
Otsu, Japan
March 23, 1960

To the Editor
TEXTILE RESEARCH JOURNAL

Dear Sir:

The Procion Black HGS procedure described in a previous paper [2] was further investigated to make clear what the staining results mean in regard to the physical or chemical structure of the fibers under observation. Particularly, washing with 30% pyridine water of stained cross section preparations was used as a criterion to judge whether or not a chemical fixation of the reactive dye had taken place onto the fiber materials. It has been proved that results obtained by the procedure should be interpreted in various ways, as discussed below.

Standard Staining Procedure

The staining solution is prepared by dissolving 10 g. of Procion Black HGS, 7 g. of urea, and 2 g. of soda ash in enough distilled water to total 100 ml. The slides carrying fiber cross sections of about 5 μ thickness are flooded with a proper amount of the above dye solution and gently heated over a small flame or on a hot plate for several minutes. Then excess dye is washed from the slides by rinsing thoroughly with water; the specimen is mounted with a suitable medium for microscopical examination.

Cellulosic Fibers

The Procion Black HGS procedure produces satisfactory staining on viscose rayons only under an alkaline pH, an essential condition for the chemical fixation of Procion dyes against the hydroxyl groups of cellulose. However, this staining bleeds readily from the cross sections upon washing with 30% pyridine water, indicating that the dye is unable to react with the fiber material when the prescribed standard procedure is applied (Figures 1 and 2). The chemical reactivity as a Procion dye appears to have no part in this case, because it was found that quite similar staining results were obtained using dye which had been elaborately hydrolyzed by boiling in an alkaline solution for several hours. It is therefore certain that the Procion Black HGS procedure, contrary to expectation, is not a chemical stain for cellulosic fibers in the same sense as the silver stain discussed in another paper [1]. The excellent differentiation obtained by the present procedure between the skin and the core in viscose rayons probably would be due to a mechanism similar to that involved in the conventional procedures employing either direct or basic dyes.

It should be added that these remarks about cellulosic fibers hold true for Vinylon fibers, too.

Staining by Procion Black HGS is markedly affected by the pH of the dye solution, but a similar pH dependence was observed for some direct dyes, though to a much lesser extent.

Protein Fibers, Natural and Regenerated

Every cross-sectional staining of protein fibers obtained by the standard procedure is fast to 30% pyridine water, so that some kind of chemical fixation is probably involved here. Naturally an alkaline pH of the dye solution is necessary for the reactive staining to occur. When applied in an acidic solution, Procion Black HGS is found to behave as an acid dye, resulting in relatively deep staining on wool, silk, casein, and even normal nylon fibers. This acidic staining, however, does not withstand pyridine water washing but bleeds off rapidly.

After alkaline hydrolysis the dye is no longer fast to washing with pyridine water, probably owing to the disappearance of the reactive groups proper to Procion dyes. On the other hand, the reactive group on the side of protein fibers seems to be diminished for the most part by acetylation with acetic anhydride. In fact, an acetylated specimen was found to show very much reduced cross-sectional staining

Fig. 1. Procion Black HGS staining according to standard procedure, demonstrating a skin-core differentiation on viscose rayon and a uniform deep stain on casein fiber.

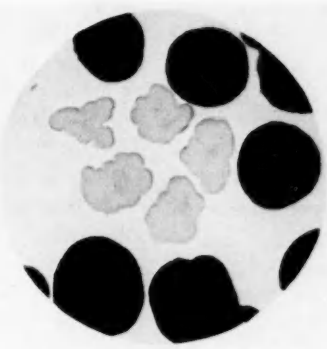

Fig. 2. Procion Black HGS staining followed by washing with 30% pyridine water on the same preparation as shown in Figure 1. The stain has been washed from viscose rayon, while casein fiber appears to be almost unaffected.

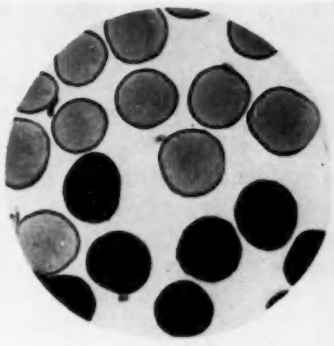

Fig. 3. Procion Black HGS staining on casein fiber followed by washing with 30% pyridine water; the much less stained cross sections are from an acetylated specimen; they are no longer capable of retaining the dye through chemical fixation.

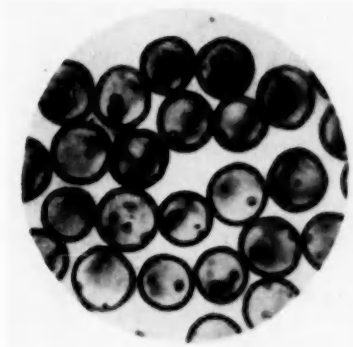

Fig. 4. Procion Black HGS staining on photodegraded nylon filament; the outermost layer as well as the areas surrounding titanium dioxide particles are particularly deeply stained.

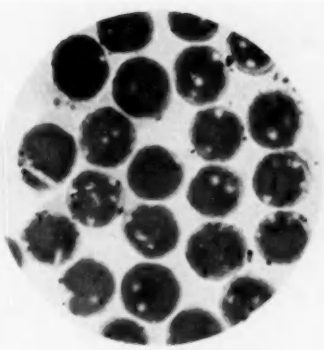

Fig. 5. Acid dye (Supranol Fast Red BB) staining on photodegraded nylon filament. This staining result is just the opposite to that shown in Figure 4.

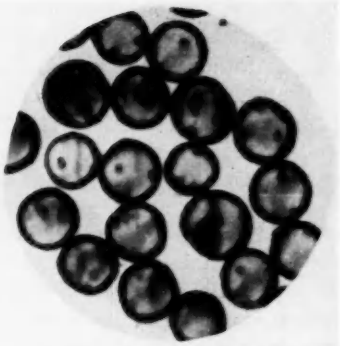

Fig. 6. Schiff's reaction on photodegraded nylon filament; this bears a close resemblance to the staining shown in Figure 4.

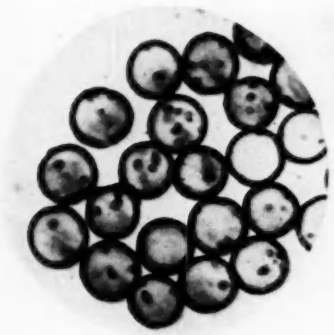

Fig. 7. Silver staining on photodegraded nylon filament; this bears a close resemblance to the results shown in Figures 4 and 6. ⟶

after washing with 30% pyridine water (Figure 3). Of course, this chemical pretreatment also results in a marked decrease of stainability with acid dyes.

It is therefore obvious that Procion Black HGS staining on protein fibers in general is dependent upon the fixative reaction of the dye with the amino groups available in the fiber materials. In an acidic

solution these amino groups may well become staining sites for acid dyes.

Nylon Filaments Degraded by Ultraviolet Irradiation

Procion Black HGS staining on photodegraded nylon filaments obtained by the standard procedure (Figure 4) is surely chemical in nature because of its fastness to 30% pyridine water washing. Staining effected under acidic pH is not fast to the same washing. However, the amino groups present in nylon filaments cannot be concerned with that chemical staining, for effects of the Procion Black HGS procedure are opposite to those of acid dyes (Figures 4 and 5). In addition, it should be remarked that neither the preinactivation of the dye by alkaline hydrolysis nor the pretreatment of the fiber specimen with acetylating agent such as acetic anhydride can affect the eventual staining results in the least. Consequently, the dye in question must be capable of going into chemical reaction with photodegraded nylon in some way other than as a simple Procion dye.

The subsequent investigation has revealed that coloring reagents for aldehydes such as Schiff's and Tollens' reagent give rise to stain reactions on the cross sections of photodegraded nylon filaments and that the results are in good agreement with those obtained by the Procion Black HGS procedure (Figure 6). In this respect the silver staining method [1], originally developed for the skin staining of viscose rayons, has also proved to be entirely the same as the above aldehyde reagents (Figure 7). Furthermore, it has been confirmed that the Procion Black HGS staining and the aldehyde color reactions run parallel to each other regarding the stainability decreases brought about by any chemical pretreatment of the specimen which can chemically modify aldehyde groups—for example, oxidation, reduction, or oximination.

It is highly probable that ultraviolet irradiation would give rise to photooxidation of nylon polyamide chains, resulting in the formation and accumulation of certain aldehydic substances as relatively stable intermediates, with which Procion Black HGS combines under weak alkaline conditions by virtue of some reactive groups, presumably amino groups, existing on its own molecules.

The Procion Black HGS procedure and the silver staining method have been found to give chemical stain reactions against the common aldehyde groups occurring on nylon filaments during photodegradation. The latter method is known to stain viscose rayon cross sections, certainly due to the reducing aldehyde groups in cellulose. It is therefore hard to understand the reason why Procion Black HGS does not react to the same aldehyde groups, in order to be fixed chemically.

An unsolved problem is that Procion Black HGS, an acid dye able to stain normal nylon filaments, does not react with the terminal amino groups under alkaline conditions. It is believed that the physical state of the dye in solution should be taken into account in order to explain these facts and the pH dependence in cellulose staining.

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KOICHI KATO

A Method of Removing the Outer Layers of Animal Fibers

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Division of Textile Physics
Commonwealth Scientific and
Industrial Research Organization
338 Blaxland Road
Ryde, N.S.W., Australia
February 15, 1960

To the Editor
TEXTILE RESEARCH JOURNAL

Dear Sir:

Little is known of the differences in properties of orthocortex and paracortex in animal fibers because of the difficulty of separation. In most coarse wool fibers (e.g., Lincoln) these components are arranged radially with the orthocortex innermost; Haly [2] has uniformly abraded such fibers in 1.5-cm. lengths to achieve separation of nearly pure orthocortex. However, for a variety of tests this length is inadequate; a device has now been developed which is capable of abrading fiber lengths of approximately 10 cm.

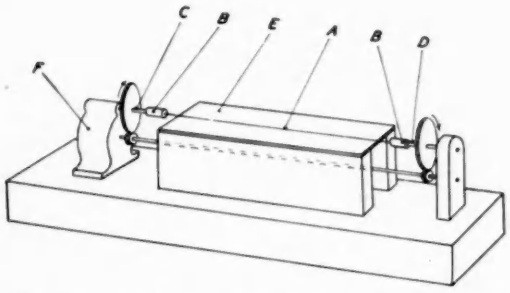

Fig. 1. Perspective view of device used for abrading fibers.

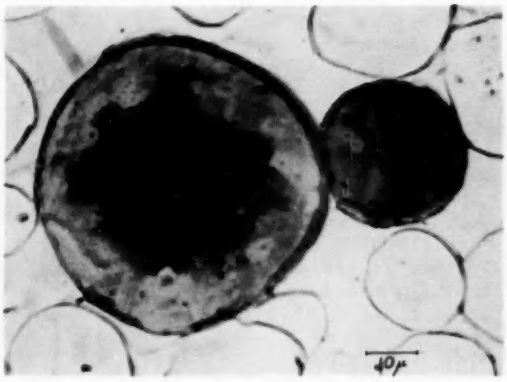

Fig. 2. Cross sections of a Lincoln wool fiber before and after abrasion. The fiber was dyed in methylene blue to show cortical asymmetry. Note that virtually all paracortex was removed and that final shape was circular.

The fiber, A in Figure 1, is cemented into two pieces of "nylex" tubing B, which are a push fit on the rotating shafts C and D. These shafts are driven by a small electric motor F, and are on the same level as the upper face of a flat sheet of glass E, the surface of which has been lightly ground. A mixture of a petroleum fraction of B.P. 200° C.–250° C. (obtained by distilling kerosene) and a fine optical abrasive powder is applied to this face. The chief function of the petroleum is to provide rapid heat transfer from the fiber to its surroundings. The fiber is rotated, sometimes at very slow rates, but in any case not exceeding approximately 500 r.p.m. A mating piece of glass 1×1 in. is then placed in contact with the fiber and plate and moved gently with combined rotary and translational motion along the length of the fiber. At regular intervals the fiber is inspected and measured under the microscope.

It has been found possible to grind lengths of up to 10 cm., finishing with a circular cross section of diameter approximately 30 μ, with a deviation from the mean diameter of $\pm 2\,\mu$. Abrading to smaller diameters results in frequent breakages.

Pure orthocortex has been obtained by this technique (see Figure 2); thus it has permitted comparison of the mechanical properties of whole fibers and the orthocortex [1]. In practice half of a single fiber is retained for the control experiment and the remainder abraded to remove paracortex before the required mechanical test is carried out. The method may also be used in studying fiber properties expected to vary with diameter. For example, Nordon [3] has investigated the kinetics of water sorption by horsehair as a function of diameter. The uptake–time relationship was obtained at several stages of the grinding of the one fiber from a diameter of 200 μ to 87 μ.

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J. W. SNAITH

A Note on the Hemicelluloses of Mesta Fiber (*Hibiscus Cannabinus*)

Technological Research Laboratories
Indian Central Jute Committee
Regent Park, Calcutta-40
India
March 28, 1960

To the Editor
TEXTILE RESEARCH JOURNAL

Dear Sir:

Mesta is a bast fiber which is used on a fairly large scale in jute manufacture. The chemical composition of the fiber is more or less similar [6] to that of jute. The structure of the hemicelluloses from mesta fiber is being studied; some preliminary results are reported here.

The composition of a sample of the fiber, analyzed by standard methods [3], is as follows: α-cellulose, 60.04%; pentosan, 16.42%; uronic anhydride, 3.87%; acetyl, 4.82%, nitrogen, 0.19%; fat and wax, 0.62%; ash, 0.69%; and lignin, 7.69%.

The fiber was cut into small pieces, defatted with alcohol: benzene mixture (1:2 wt./wt.) and delignified by the sodium chlorite method [4]. The holocellulose thus obtained was treated at room temperature with 10% NaOH (wt./vol.; liq. ratio 1:10) for 2 hr. The mixture was then filtered through a sintered glass funnel. The cold filtrate was neutralized with glacial acetic acid and hemicellulose I was precipitated by adding an equal volume of ethanol. It was filtered and to the clear filtrate more ethanol was added to make the concentration of alcohol 80% (vol./vol.); this precipitates hemicellulose II. Both hemicellulose I and II were purified by treatment with normal ethanolic hydrochloric acid, then washed with ethanol and dried. They were obtained as white amorphous powder and represent 14% and 1% respectively of the dry weight of the fiber. The analytical data are given in Table I.

Hemicellulose I and II were hydrolyzed with normal sulfuric acid on a boiling water bath for 10 hr. Paper chromatographic examination of their hydrolysates revealed the presence of both acidic and neutral sugars. With the solvent system ethyl acetate: acetic acid: formic acid: water (18:3:1:4), the acidic components corresponded chromatographically to 4-O-methyl-D-glucuronic acid and 2-O-(4-O-methyl-α-D-glucuronosido)-D-xylose similar to that from jute [1, 5, 8]. The neutral sugars were mainly xylose together with minor quantities of galactose, glucose, mannose, and rhamnose in hemicellulose I and, in addition, arabinose in hemicellulose II. They were separated in Whatman No. 1 chromatography paper using the solvent system butanol: pyridine: water (10:3:3) and estimated by use of the Somogyi copper reagent [7]. The proportion of sugars present in both hemicellulose I and II (calculated as percent of mole fraction) is given in Table II.

The hydrolysate of hemicellulose I was neutralized with $Ba(OH)_2$ and treated with Amberlite IR-120 cation exchange resin. The concentrated solution was added to the top of a column containing IR-45 (OH) anion exchange resin and eluted with water. Evaporation of the eluate gave crystalline D-xylose, mp. and mixed mp. 144° C. and $[\alpha]_D^{30} + 18°$ (c; 1.5 in water) after recrystallization from aqueous alcohol. The dibenzilidine dimethyl acetal derivative [2] had mp. and mixed mp. 210° C. and $[\alpha]_D^{27} - 11.7°$ (c; 1.5 in chloroform).

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TABLE I

	Pentosan, %	Uronic anhydride, %	Methoxy, %	Eq. wt.	$[\alpha]_D^{30}$
Hemicellulose I	79.57	17.98	3.07	1093	−47.03° (c; 0.69 in *N* NaOH)
Hemicellulose II	46.88	14	3.14	1117	−30.35° (c; 0.5 in *N* NaOH)

TABLE II

	Galactose	Glucose	Mannose	Arabinose	Xylose	Rhamnose
Hemicellulose I	0.68	1.38	1.15	—	95.68	1.08
Hemicellulose II	8.64	17.26	6.22	6.02	57.67	4.2

Some Physical Observations on Roselle

Department of Fundamental Physics
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Regent Park, Calcutta-40, India
March 14, 1960

To the Editor
TEXTILE RESEARCH JOURNAL

Dear Sir:

Dependency on jute fiber from India and Pakistan has led many countries to cultivate large numbers of fiber plants, among which three or four, such as *Hibiscus cannabinus* (Bimli jute, kenaf, or mesta), *Hibiscus sabdariffa* (roselle), and *Urena lobata* (ararium fiber), are trying to compete as jute substitutes, to some extent effectively. As roselle is mostly used as a jute blend it is necessary to study the physical properties of both fiber and yarn.

In this article the author has studied, in the form of single filaments, the gravimetric fineness and breaking load at different test lengths and consequently the substance strength of roselle fiber in order to give comparative data with that of ramie [2], jute, and allied fibers [4]. These findings will also supplement the findings of Bandopadhyay and Bose [1] and add one more item to the list of Meredith [3].

Three roselle samples of known yarn qualities were selected for study.

	J5905	J5906	J6653
Quality ratio	76	62	47
Weight CV, %	26	30	26

For fiber fineness a semi-microbalance and for breaking load a Baer Single Fiber Tester were used. Tests were carried out at test lengths ranging from 1 cm. to 10 cm. at intervals of 1 cm. For each test length 150 single filaments were separated randomly from a properly combed sample and cut to a size 3 cm. greater than the actual test length.

After the fineness value was ascertained, each fila-

TABLE I

Test length, cm.	Filament mass, den.			Breaking load, g.			Substance strength, g./den.			
	J5905	J5906	J6653	J5905	J5906	J6653	J5905	J5906	J6653	Mean
1	25.20	24.12	28.71	123.4 ± 5.59	92.9 ± 4.80	124.1 ± 5.33	4.90	3.85	4.32	4.36
2	24.03	28.17	26.82	101.3 ± 4.61	97.6 ± 4.63	98.6 ± 4.51	4.22	3.46	3.68	3.79
3	24.75	24.75	24.66	92.7 ± 4.59	73.5 ± 4.07	80.5 ± 4.07	3.75	2.97	3.26	3.33
4	25.83	35.19	24.75	85.9 ± 4.57	107.7 ± 5.03	75.4 ± 3.96	3.33	3.06	3.05	3.14
5	28.35	33.93	26.46	93.7 ± 4.34	105.5 ± 5.08	68.5 ± 3.31	3.31	3.11	2.59	3.00
6	24.84	25.47	31.95	71.8 ± 3.96	62.6 ± 3.28	86.1 ± 4.22	2.89	2.46	2.69	2.68
7	25.02	34.56	29.97	62.7 ± 3.19	95.5 ± 4.59	71.1 ± 3.78	2.51	2.76	2.37	2.55
8	30.60	33.03	32.22	73.1 ± 2.47	77.4 ± 3.68	71.8 ± 3.54	2.39	2.34	2.23	2.32
9	38.70	31.05	34.38	86.3 ± 3.43	69.6 ± 3.24	80.1 ± 3.41	2.23	2.24	2.33	2.27
10	29.25	32.85	26.55	57.4 ± 3.26	61.2 ± 3.30	46.6 ± 2.66	1.96	1.86	1.76	1.86
Mean	27.66	30.31	28.65							

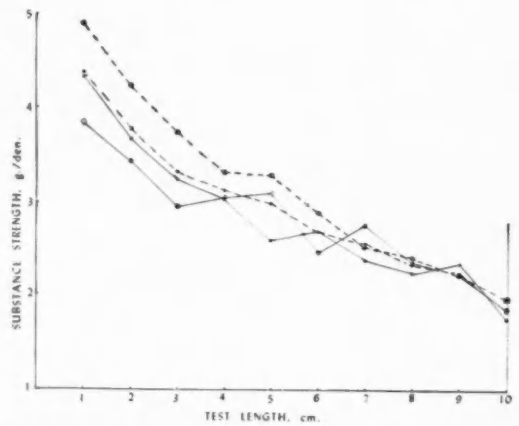

Fig. 1. Length vs. strength. o- - -o, J5905; •——•, J6653; o——o, J5906; •- - -•, mean.

ment was tested for breaking load. Tests were conducted in an atmosphere of 65 ± 3% RH; Table I gives the results.

The substance strength variations are graphically shown in Figure 1.

From the data it is evident that J5905, having the highest substance strength and lowest mass per unit length, will have the best yarn qualities, high QR, and low CV. In keeping with the usual relation of filament coarseness with higher yarn CV, J5906, being the coarsest, gives the highest yarn CV. Presumably the decrease in strength of J5906 has been influenced by its coarseness.

The generally accepted exponential nature of the curve is more or less well maintained in all the three samples. A smoother curve is, however, obtained if the mean of the three is plotted against test length. Excepting ramie, substance strength of roselle can be compared favorably to that of other bast fibers, but it is comparatively coarser than jute.

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Polymerization of Acrylonitrile from the Vapor Phase onto Cotton by Gamma Irradiation

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February 25, 1960

To the Editor
TEXTILE RESEARCH JOURNAL

Dear Sir:

Recently Arthur, Demint, McSherry, and Jurgens [1], in a Letter to the Editor, reported the polymerization of acrylonitrile on cotton yarn by gamma radiation. Solutions of the monomer in aqueous $ZnCl_2$ were used for impregnating the samples prior to irradiation.

In a forthcoming paper, it is shown that acrylonitrile, as well as other volatile vinyl monomers, may be added to fibrous polymers from the vapor phase by gamma radiation. The presence of water is essential.

In the system cotton cellulose–acrylonitrile vapor–water vapor, a uniform product of a remarkably soft hand is obtained even at a polyacrylonitrile content of as much as 25%. The amount of deposition of polymer may be controlled by either the concentration of acrylonitrile in the vapor phase or by the dose of radiation.

In the vapor-phase addition of monomers, the fiber substrate appears to influence the amount of polymer fixed at any given dose of radiation. The relatively unstable fibers show the greatest addition.

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